Investigation into the suitability of spring triticale (×*Triticosecale* Wittmack) for bio-ethanol production in the Western Cape

by

Yuriy Vadimovich Tsupko

Thesis presented in partial fulfilment of the requirements for the degree of Master of Science in Agriculture (Genetics)

at

Stellenbosch University

Department of Genetics

Faculty of AgriScience

Supervisor: Willem C. Botes

December 2009

Declaration

By submitting this thesis electronically, I declare that the entirety of the work contained therein is my own, original work, that I am the owner of the copyright thereof (unless to the extent explicitly otherwise stated) and that I have not previously in its entirety or in part submitted it for obtaining any qualification.

December 2009

Abstract

In the Western Cape small grain cereals, triticale (×*Triticosecale* Wittmack ex A. Camus) in particular, appear to be among the most promising starch-carrying raw materials for the production of bio-ethanol. A core group of cultivars and lines from the Stellenbosch University Plant Breeding Laboratory spring triticale breeding programme were subjected to initial testing for the purpose of ethanol production. They underwent multi-location field-testing across six (season 2006–2007) and nine (season 2007–2008) locations representing the Western Cape cereal production area.

Climatic conditions during the study were characterised as generally favourable, especially in the 2007 season. During the season, trials were visited in order to make *in situ* observations. Disease susceptibility was given specific attention. After harvesting, grain yield ($kg.ha^{-1}$), test weight ($kg.HL^{-1}$), total starch content in whole grain (%), amylose/amylopectin ratio, protein content (%), ethanol output ($L.tonne^{-1}$) and ethanol yield ($L.ha^{-1}$) were analysed.

Near infra-red reflectance spectroscopy calibration models were developed for moisture and starch contents. The best calibration based on whole grain spectra for moisture content had RPD = 1.691, $R^2 = 0.657$ and SEP = 0.271%, and for starch content RPD = 1.646, $R^2 = 0.634$ and SEP = 1.356%. Calibrations developed from milled grain showed better results for moisture content RPD = 2.526, $R^2 = 0.843$, SEP = 0.182%, and for starch content RPD = 1.741, $R^2 = 0.673$, SEP = 1.277%. These calibrations are suitable for rough screening of samples.

In the 2006 season, starch yield was highly positively correlated with grain yield ($R^2 = 0.988$, $P < 0.001$). Both starch yield and grain yield were positively correlated with days to heading ($R^2 = 0.533$ and $R^2 = 0.556$, respectively; $P < 0.001$).

The 2007 season was characterised by a generally higher starch yield (2952–3142kg.ha^{-1}, 95%CI) compared to the 2006 season (2077–2315kg.ha^{-1}, 95%CI). Starch yield was strongly positively correlated with grain yield ($R^2 = 0.975$, $P < 0.001$). Test weight demonstrated weak positive correlation with ethanol yield ($R^2 = 0.238$, $P < 0.01$) and grain yield ($R^2 = 0.279$, $P < 0.001$). Mean ethanol output ranged between 466–477L.tonne^{-1} at the 95%CI. Ethanol output was demonstrated to be more dependent on starch and other polysaccharides accessibility to enzymatic digestion than on the total starch content as such. The best lines for ethanol output in the 2007 season were G2, D3 and H2 for the Swartland region, and D3, G2 and D1 for the Overberg region.

The best triticale lines under investigation showed their potential from a biological point of view to be a suitable crop for ethanol production in the Western Cape, with the achieved ethanol yield ranging between 2446–2625L.ha^{-1} at the 95%CI. For the Swartland region the best genotypes for ethanol yield were D1, H1 and D2, and for the Overberg H1 and G2. The 23 best lines were selected from the elite and senior blocks, and then used for the establishment of a recurrent mass-selection pre-breeding block.

Opsomming

In die Wes-Kaap is kleingrane, meer spesifiek korog (×*Triticosecale* Wittmack ex A. Camus), van die mees belowende styseldraende rou-materiale vir die produksie van bio-etanol. ‘n Kern versameling van kultivars en telerslyne van die Universiteit van Stellenbosch se Planteteeltlaboratorium se lente korogteeltprogram is blootgestel aan aanvanklike toetsing met die doel om etanol produksie te meet. Die materiaal het veldtoetsing ondergaan oor verskeie lokaliteite gedurende die 2006–2007 (ses lokaliteite) en 2007–2008 (nege lokaliteite) seisoene wat verteenwoordigend was van die Wes-Kaapse produksie gebied.

Klimaatstoestande gedurende die studie kan beskryf word as gunstig, veral gedurende die 2007 seisoen. Gedurende die groeiseisoen is proeflokaliteite gereeld besoek ten einde *in situ* observasies te kon maak, siektevatbaarheid het veral aandag geniet. Na die oes van proewe was graanopbrengs ($kg.ha^{-1}$), hektolitermassa ($kg.HL^{-1}$), totale-styselinhoud in heelgraan (%), amilose/amilopektien-verhouding, proteïeninhoud (%), etanolopbrengs ($L.ton^{-1}$) en etanolopbrengs per hektaar ($L.ha^{-1}$) gemeet.

Naby-infrarooispektroskopie kalibrasies was ontwikkel vir vog- en styselinhoud. Die beste kalibrasies vir heelgraan voginhoud het ‘n RDP = 1.691, R^2 = 0.657 en SEP = 0.271% en vir styselinhoud RPD = 1.646, R^2 = 0.634 en SEP = 1.356% opgelewer. Die kalibrasies gebaseer op meel was aansienlik beter vir voginhoud RPD = 2.526, R^2 = 0.843 en SEP = 0.182%, sowel as vir styselinhoud RPD = 1.741, R^2 = 0.673 en SEP = 1.277%. Die kalibrasies is bruikbaar vir aanvanklike sifting van monsters.

Gedurende die 2006 seisoen het styselinhoud en graanopbrangs ‘n baie hoë korrelasie (R^2 = 0.988, P <0.001) getoon. Beide stysel- en graanopbrengs was positief gekorreleerd met dae tot aar (R^2 = 0.533 en R^2 = 0.556; P <0.001).

Die 2007 seisoen is gekenmerk deur ‘n hoër styselopbrengs (2952–3142kg.ha^{-1}, 95%VI) teenoor die 2006 seisoen (2077–2315kg.ha^{-1}, 95%VI). Styselopbrengs was positief gekorreleerd met graanopbrengs (R^2 = 0.975, P <0.001). Hektolitermassa het swak korrelasie getoon met etanolopbrengs (R^2 = 0.238, P <0.01) en graanopbrengs (R^2 = 0.279, P <0.01). Gemiddelde etanolopbrengs het gewissel tussen 466–477L.ton^{-1} by 95%VI. Data het aangedui dat etanolopbrengs meer aangewese is op stysel en ander polisakkariedverbindings se ensiematiese toeganklikheid eerder as totale stysel aanwesig. Die beste lyne wat etanolopbrangs betref in 2007 was G2, D3 en H2 vir die Swartland en D3, G2 en D1 vir die Overberg.

Van die koroglyne wat deel was van die ondersoek het goeie potensiaal getoon, uit ‘n suiwer biologiese oogpunt, as gewas vir die produksie van etanol in die Wes-Kaap met ‘n gerealiseerde etanolopbrengs in die omgewing van 2446-2625L.ha^{-1} by 95%VI. In die Swartland was die beste genotipes D1, H1 en D2 en in die Overberg H1 en G2. Die beste 23 lyne is geselekteer uit die elite en senior telingsblokke en aangewend in die vestiging van ‘n herhalende-seleksie voortelingsblok.

Acknowledgements

The author wishes to acknowledge contribution of the following individuals and organisations throughout the years of study without whom completion of this work would be impossible:

My scientific supervisor WILLEM BOTES (head of the Plant Breeding Laboratory, Department of Genetics, Stellenbosch University, Republic of South Africa) for the trust he has put into me, for financial support, suggestions and encouragement given, tolerance and patience throughout the duration of this project.

Staff and contract workers of the Stellenbosch University Plant Breeding Laboratory for their support and friendly attitude. Special thanks go to CHARLES TOUTIE, HENZEL SAUL and ANDRE JULIUS for their help in the greenhouse and in the field trials, MELANIE LEO and CORRIE OOSTHUIZEN for their help with moisture content determination, and ALETTA EKSTEEN for her help with ethanol output determination.

Dr MARENA MANLEY (Department of Food Science, Stellenbosch University, Republic of South Africa) for her friendliness, for introducing me into and teaching of the near infra-red spectroscopy methods, for granting access to her library, as well for the supply of protein and particle size index raw data.

Staff of the International Office of the Stellenbosch University for help with my registration and visa renewals.

RITHA WENTZEL (Agro-Climatology, ARC Institute for Soil, Climate and Water, Stellenbosch, Republic of South Africa) for the supply of precipitation, maximum and minimum temperatures raw data.

My former chiefs at the Institute of Plant Production n.a. V.J. Yurjev of the UAAS, Ukraine: Dr GENNADIY SHCHIPAK (head of the Department of Triticale Breeding) and Dr VICTOR RYABCHOUN (head of the National Centre for Plant Genetic Resources of Ukraine) for the provision of literature sources regarding history and current state of triticale breeding in countries of the former Soviet block.

Dr SVETLANA DIDENKO (Laboratory of Grain Quality, Institute of Plant Production n.a. V.J. Yurjev of the UAAS, Ukraine) for the help with Russian and Ukrainian literature sources regarding starch research.

Dr FREDERICK STODDARD (Department of Applied Biology, University of Helsinki, Finland), Dr ROBERT GRAYBOSCH (USDA-ARS, University of Nebraska, Lincoln, NE, USA) and Dr GRANT MCLEOD (Agriculture and Agri-Food Canada, Indian Head, SK, Canada) for meaningful email conversations regarding creation of high-amylopectin and high-amylose triticale.

Developers of excellent freeware software packages, which I employed for my data analysis.

My landlord RUDI STEGMÜLLER and his wife JOHANNA for their magnanimous generosity in providing me with virtually free accommodation throughout years of my study in South Africa. Without it, I could hardly cut and contrive.

My friends at the Helderberg College (Somerset West, Republic of South Africa), particularly members of the Silverleaf congregation of the Seventh-day Adventist Church for their fellowship, prayers, spiritual support and real help in my various needs. They made me feel at home in this distant land; may Luke 6:38 be fulfilled in their life. I would like to specifically mention YVETTE SPARROW, AUDREY KING, JEANEY ROSE, ANDRE MALHERBE, JACO SCHOLTZ, as well as TANYA MEDINSKI (Stellenbosch University) for a sacrifice of their time in answering my questions regarding English language use and help with proofreading of various parts of the manuscript. My friends MARCOS CRUZ and his wife DEIDRE WARELEY-CRUZ, CONRAD ZYGMONT and JOHN CHIPIMPI for help with transport when I needed it. Special thanks go to HELEN GONCHAROVA for being my only Russian-speaking friend during most of the duration of my stay in South Africa, for her moral support and unselfish spirit in sharing of her daily bread with me.

My mother LIUBOV TSUPKO for her constant prayers and encouraging emails, for interest in my life, for bringing me up, giving me education, for her support in developing my abilities in a way that I could become who I am and get where I am today; without her, it would not have been possible.

My Creator and Redeemer JESUS CHRIST, who led my way into South Africa, sustained my existence and health throughout the days of my life. All the glory goes to Him, who remains the only Faithful and True One in spite of all circumstances.

Yuriy Tsupko
tsupko@gmail.com

Table of Contents

List of Figures

List of Tables

List of Addendums

List of Abbreviations

3-PGA – 3-phosphoglyceric acid
α-AMY – α-amylase (alpha-amylase)
AAC – apparent amylose content
AACC – American Association of Cereal Chemists
AAQ – auto-amylolytic quotient
AD – amphidiploid
ADP-Glc – adenosine diphosphate glucose (= ADP-glucose)
ADP-glucose – adenosine diphosphate glucose
AGPase – ADP-glucose pyrophosphorylase
AMG – amyloglucosidase
AMMI – additive main effects and multiplicative interaction
ANN – artificial neural network
ANOVA – analysis of variance
AOAC – Association of the Official Analytical Chemists
ARC – Agricultural Research Council
ATP – adenosine triphosphate
BG – β-glucan (beta-glucan)
CIMMYT – Centro Internacional de Mejoramiento de Maiz y Trigo
(Spanish for "International Maize and Wheat Improvement Centre")
Con A – concanavalin A
DBE – starch-debranching enzyme (= SDE)
DDGS – distillers dried grains with solubles
DM – dry matter
DMSO – dimethyl sulphoxide
DNA – deoxyribonucleic acid
DP – degree of polymerisation (also called average chain length)
DSC – differential scanning calorimetry
DWB – dry weight basis
EMS – ethyl methanesulfonate
EO – ethanol output
EY – ethanol yield
FS – fermentable substance
FT-NIR – Fourier transform near infra-red
G×E – genotype-by-environment [interaction]
GAC – grain-amylose content
GBSS – granule-bound starch synthase
GLC – gas-liquid chromatography
GM – genetic modification; genetically modified
GOPOD – glucose oxidase/peroxydase
GWD – α-glucan water dikinase
HAC – hierarchical agglomerative clustering
HL – hectolitre
HPLC – high performance liquid chromatography
HTC – hydro-thermal coefficient
HTF – high total fermentable
ICC – International Association for Cereal Science and Technology
IPCA – interaction principal component axis
ITYN – international triticale yield nursery
LT – long-term

MARS – marker-assisted recurrent selection
MCPA – 2-methyl-4-chlorophenoxyacetic acid
MLFT – multi-location field-testing
MLR – multiple linear regression
MOPS – 4-Morpholinepropanesulfonic acid
MSC – multiplicative signal correction
NIRS – near infra-red reflectance spectroscopy
NITS – near infra-red transmittance spectroscopy
NSP – non-starch polysaccharides
PC – principal component
PCA – principal components analysis
PCR – polymerase chain reaction
Pi – inorganic orthophosphate
PIN-a, PIN-b – puroindolin isoforms of friabilin proteins
PLS – partial least squares
PLSR – partial least squares regression
PSI – particle size index
QTL – quantitative trait loci
R^2 – coefficient of determination
RAP – relative ability of prediction
RCBD – randomised complete block design
REML – restricted (or residual) maximum likelihood
REO – relative ethanol output
RMSEP – root mean square error of prediction
RNA – ribonucleic acid
RNAi – ribonucleic acid interference
RPD – relative predictive determinant, or the ratio of performance to deviations
RPM – rotations per minute
RSA – Republic of South Africa
RuBisCO – ribulose-1,5-bisphosphate carboxylase-oxygenase
SAC – starch-amylose content
SBE – starch-branching enzyme
SD – standard deviation
SDF – soluble dietary fibre
SEC – 1. size exclusion chromatography; 2. standard error of calibration
SECV – standard error of cross-validation
SEP – standard error of performance
SGP – starch granule-bound protein
SRC – Spearman' rank correlation
SS – starch-synthase
SSF – simultaneous saccharification and fermentation
SU-PBL – Stellenbosch University Plant Breeding Laboratory
SuSy – sucrose-synthase
TDP – ten-day period
TILLING – targeting induced local lesions in genomes
UAAS – Ukrainian Academy of Agricultural Science
UDP-glucose – uridine diphosphate glucose
USA – United States of America
USDA-ARS – United States' Department of Agriculture - Agricultural Research Service
USSR – Union of Soviet Socialist Republics
VHG – very-high gravity [fermentation]
VWE – viscosity of the water extract

Chapter 1: Introduction

"Control oil and you control nations;
control food and you control the people"
Henry Alfred Kissinger,
the former USA secretary of state, 1974

1.1. STUDY BACKGROUND

Cereal producers of South Africa's Western Cape Province have been struggling to maintain profit margins due to several factors. The identification of alternative, more profitable and stable markets for cereals is therefore much desired. The South African government is encouraging the establishment of a bio-fuels industry in the hope that it will contribute to job creation and the emergence of small-scale farmers, as well as to a lowering of greenhouse gas emissions, diversification of fuel supplies and a reduction of South Africa's crude oil import bill (DME RSA, 2006, 2007). In early 2006, a Biofuel Task Team was launched in the Western Cape to investigate the viability of establishing a bio-ethanol production plant for the region as well as the role that Ethanol Africa, Grain South Africa, and the South African Petroleum Industry Association can play (GRAIN SA, 2006). A successful bio-ethanol industry in the Western Cape could potentially assist producers in returning to profitability, and could potentially increase employment in rural areas by promoting the use of first generation bio-fuels (ANONYMOUS, 2006; DME RSA, 2007).

Biofuels are generally seen as a substitute for fossil fuels and a possible solution for combating global warming and climate change. However, there some disputes regarding the energy balance of biofuels, their renewability and their long-term effect on society, the environment and the economy (PIMENTEL, 2003, 2006; PATZEK, 2004, 2006a-c; HENCKE, KLEPPER & SCHMITZ, 2005; PATZEK *et al.*, 2005; PIMENTEL & PATZEK, 2005; ANONYMOUS, 2006; CAIRNS, 2006; PATZEK & PIMENTEL, 2006; BAILEY, 2007; REIJNDERS & HUIJBREGTS, 2007; REICHARDT, 2007; ANONYMOUS, 2008; DEAT RSA, 2008; KEYZER, MERBIS & VOORTMAN, 2008; SAWYER, 2008; SEARCHINGER *et al.*, 2008; STALEY & BRADLEY, 2008; SYLVESTER-

BRADLEY & KINDRED, 2008; VAN WEY, 2009). In spite of the above issues, the topic of the production of ethanol and its co-products from small grain cereals, triticale (×*Triticosecale* Wittmack ex A. Camus) in particular, still merits exploration from the plant breeding point of view.

In the Western Cape small grain cereals – namely wheat (*Triticum aestivum* L.), triticale, barley (*Hordeum vulgare* L.) and oats (*Avena sativa* L.) – appear to be among the most promising starch-carrying raw materials for the production of ethanol and a broad range of other products for various industrial implementations including being used as a possible partial substitute (oxygenate) of the conventional liquid fossil fuel counterparts that are currently used in the petrochemical industry (POLMAN, 1994; FORWARD, 1994; SLATTERY, KAVAKLI & OKITA, 2000; KIM & DALE, 2004; KOUTINAS, WANG & WEBB, 2004). Triticale can provide an ideal raw material for the formation of a generic fermentation feedstock for ethanol production, as it contains all the required nutrients to induce microbial development and final product formation. Although the crop was initially developed for bread making and certainly possesses untapped potential for such use, triticale is not used for food production in South Africa, and thus is not limited by the same restrictions and regulations as wheat and other traditional food cereals in terms of other industrial uses and its genetic modification (GRESSEL, 2008).

In many regions of the world, triticale has found a definite role as a crop for low growing cost systems. It can be grown on marginal soils (e.g. acidic, alkaline, light and drought-prone) and in 2nd and 3rd positions in the cereal rotation, which are less suited for wheat (VARUGHESE, PFEIFFER & PENA, 1997; KARPENSTEIN-MACHAN & SCHEFFER, 1998; OVERTHROW & CARVER, 2003; EREKUL & KOHN, 2006; DAVIS-KNIGHT & WEIGHTMAN, 2008). Triticale's better disease resistance compared to wheat or barley is a major advantage, which makes triticale particularly suited to organic farming systems. It has lower input requirements (particularly lower nitrogen requirement levels) with considerably less management operations compared to wheat, giving it both economic and environmental advantages. It has potential for use

as a whole crop where high quality, high dry matter yet low-cost feed is required for on-farm feeding. This especially applies to mixed farms. Triticale is, therefore, a crop that is particularly suited for low yield potential environments, e.g. acid or drought-prone soils, and where disease pressure is high, which is the case in the Western Cape Province. Given that triticale is highly adaptable, can be grown with reduced levels of inputs, and can outperform other cereals on marginal land, it will have a competitive advantage over other cereals in terms of productions costs (HACKETT & BURKE, 2004).

Since the early 1970s, the Stellenbosch University's Department of Genetics has been instrumental in developing superior triticale cultivars for use within the Winter Rainfall Region of South Africa (PIENAAR, 2006). So far, Stellenbosch University is the only institution in Africa with a dedicated spring triticale-breeding programme.

1.2. PURPOSE OF THE STUDY

Major cereal crop plants (except maize *Zea mays* ssp. *mays* L.) have been bred primarily for food or feed production, and never had selection pressure applied to optimise their traits for industrial bio-fuel. Some other industrial crops, namely sugar cane (*Saccharum* spp.) and sugar beet (*Beta vulgaris* L.) have been bred specifically for high-sugar yields. They can be used as raw material for ethanol production. However, both above-mentioned crops are unsuitable for industrial propagation in the Western Cape region, considering climatic conditions. Breeders need information from industry to be most efficient in creating the best adapted raw material for industrial purposes. Unfortunately, little data has been published internationally on the production of ethanol from small grain cereals (especially wheat and triticale) and their breeding for this purpose. This is understandable, because the topic of bio-fuels is relatively new, and historically worldwide breeding efforts were aimed at traits of interest that are unrelated to ethanol production objectives. Most of contemporary

research on bio-ethanol production from cereals is focused on maize, a crop that is not suited for the environmental conditions of the Winter Rainfall Region of South Africa. Additionally, in many cases research is not made public due to engrossed intellectual property rights considerations, especially those involved in the industrial processes (GLENNA & CAHOY, 2009). Thus far, results regarding ethanol output, yield and auto-amylolytic quotient (AAQ) are only available for a small number of triticale cultivars. They do not allow estimating genotypic variation, which is a prerequisite for breeding for improved ethanol production. However, genetic components for higher ethanol yield do exist, although have never specifically been targeted in the past. Thus, there appears to be a considerable opportunity to evaluate and optimise field crops, triticale in particular, for use in bio-energy strategies.

1.3. RESEARCH QUESTIONS

Plant breeders are expected to start working on the development of cultivars that would be better adapted to the demands of ethanol production industry. The main purpose of this study was specifically to evaluate spring triticale as possible feedstock for bio-ethanol production in the Western Cape region of South Africa. To be successful in plant breeding it is crucial to know the necessary characteristics of a cultivar for production of bio-ethanol. Only traits that have high heritability (having consistency over the years) should be used. The basic questions for the plant breeder are: the dimension of the expected market for a new cultivar; characterisation of the breeding trait 'suitability for production of bio-ethanol'; which crops will ultimately be used for production of bio-ethanol; and whether there is an association to other breeding traits, e.g. the industrial production of starch. All these questions are important and valid, and are waiting to be answered in due order, considering availability of resources, time and necessary expertise. The scope of the research in this study was limited to the following areas:

- optimisation and deployment of testing protocols for traits of interest;

- evaluation of existing South African triticale cultivars and advanced breeding lines;
- establishment of 'triticale-for-bio-ethanol' pre-breeding block for the Western Cape environmental conditions.

The following section provides necessary theoretical and practical background for more detailed clarification of questions addressed in our study.

1.4. RESEARCH METHOD: AIM AND OBJECTIVES

1.4.1. Aim

The study was focused on two aspects concurrently. The first is the *multi-location field testing* of existing cultivars and advanced breeding material from the Stellenbosch University Plant Breeding Laboratory (SU-PBL)[*] triticale breeding programme over potential production areas in the Western Cape Province, with a strong focus on areas that have not been economically viable in recent years for bread wheat production. In several locations, trials have been grown over years, which were designed to show the genetic influence of traits of interest as well as to reveal whether selections and breeding effort could be successful. The data stemming from this was analysed to establish a better understanding of current potential of triticale for bio-ethanol production in the abovementioned region.

The second part was focused on establishing a pre-breeding effort by *optimising testing protocols* for rapid screening of ethanol production potential, *identifying agronomic characteristics* coupled with potential increases in ethanol yield, and *quantifying levels of genetic variability* regarding desired traits for ethanol production in local germplasm, as well as *establishing of a pre-breeding block* where the traits of interest linked with high ethanol yield are exploited.

[*] Plant Breeding Laboratory, Department of Genetics, Stellenbosch University, Private Bag X1, Matieland 7602, South Africa.

1.4.2. Objectives

For the duration of the project, two consecutive seasonal runs of field and laboratory tests were performed, which are briefly described below (for more details see **Chapter 3: Materials and methods**).

Multi-location field-testing (MLFT). During early 2007 and the following 2008 season, a core group of cultivars and lines from the SU-PBL spring triticale breeding programme were subjected to initial testing of grain for starch content, fermentable sugar levels and ethanol yield. The pedigrees of the plant material involved are given in Tables 3.1.2 – 3.1.4. These cultivars and lines underwent MLFT across six (season 2006) and nine (season 2007) locations representing the Western Cape cereal production area (Table 3.1.1; Figures 3.1.2 and 3.1.3). Each location consisted of 20 entries in four (2006) and three (2007) repetitions. During the season, trials locations were visited on a regular basis in order to make *in situ* observations and data recording of agronomic characters such as plant height, date of heading, etc. Disease susceptibility was given specific attention, being such a yield-limiting factor. Each plot was harvested *in toto*; the seeds cleaned and kept for subsequent analysis.

Laboratory analysis and data collection. Laboratory analysis was performed and data recorded for the following traits: grain yield ($kg.ha^{-1}$); test weight ($kg.HL^{-1}$); total starch content in whole grain (% dry matter) by a α-amylase/amyloglucosidase method (AACC Method 76-13; McCleary, Gibson & Mugford, 1997); amylose/amylopectin ratio by a concanavalin A method (Gibson, Solah & McCleary, 1997); protein content (%) by a near infra-red reflectance spectroscopy method; ethanol output with (+E) and without (–E) the addition of technical enzymes (measured in $L.tonne^{-1}$; Senn & Pieper, 2001); AAQ as the ratio between ethanol output –E and +E (%); and ethanol yield +E ($L.ha^{-1}$). Statistical analysis was performed with CropStat for Windows 7.2.2007.3 (International Rice Research Institute, Metro Manila, Philippines), KyPlot 2.0 Beta 15 32-bit (Koichi Yoshioka, Japan) and GGEbiplot Pattern Explorer 6.0 (Weikai Yan).

Near infra-red reflectance spectroscopy (NIRS) analysis. Triticale samples reflectance spectra were measured by means of NIRS using a BÜCHI NIRLab N-200 on whole grain and milled grain samples. NIRS calibration models for starch and moisture content were developed using software The Unscrambler 9.2 (CAMO, Norway), with the aim to evaluate their robustness and performance as a rapid surrogate screening method to possibly be used instead of wet-chemical analytical methods for prediction of the traits parameters.

Establishing of a pre-breeding block. Cultivars and advanced lines which showed high-starch and high-ethanol yield potential were selected and inter-crossed, founding a core of a specialised ethanol-breeding nursery.

1.5. EXPECTED RESULTS

Analytical protocols tested and optimised for the breeding programme purposes. NIRS-based calibrations developed and tested for the traits of interest. Genetic diversity of the available germplasm explored and evaluated for the subsequent breeding of cultivars for increased ethanol production. Promising high-starch and high-ethanol yielding lines used in establishing of a recurrent selection scheme in bio-ethanol pre-breeding nursery. Field-testing and observations *in situ* have given a preliminary idea of better-suited triticale cultivars, and have indicated which production areas could be economically viable for propagation of triticale cultivars for bio-ethanol production purposes. It contributed data towards a bigger project on the investigation of the suitability of cereal crops for bio-ethanol production in the region. Research results presented at plant breeding conferences as well as published in peer-reviewed journals.

1.6. STRUCTURE OF THE THESIS

The following **Chapter 2: Literature Review** contains a review of background literature that gives necessary framework for the study approach and

methodology, beginning with the origin of triticale, history of its development and uses. Topics related to structural and fractional organisation of polysaccharide complex of cereal plants, its synthesis, genetic regulation, technological properties, and methods of determination are described. Objectives and approaches to breeding of high ethanol yielding cultivars were reviewed.

Chapter 3: Materials and methods describes the technical part of the study; outlines the methodological framework and introduces materials, equipment, methods and software used.

Chapter 4: Results and discussion presents raw data collected; reports analysis of the work done in implementation and optimisation of the testing protocols for rapid screening of ethanol production potential; deals with the identification of agronomic characteristics coupled with potential increases in ethanol yield, genetic variability levels' quantification and their use in a new high-starch, high-ethanol breeding nursery establishment.

Chapter 5: Conclusions and recommendations sums up the study and gives final conclusions and recommendations for further work.

Chapter 2: Literature review

2.1. TRITICALE – ITS ORIGIN AND USES

Triticale (accepted taxonomic name ×*Triticosecale* Wittmack ex A. Camus – see STACE, 1987; SORENG, 2003), contrary to the rest of agricultural crops, is created artificially by the intercrossing of wheat (*Triticum* spp.) and rye (*Secale* spp.) with the subsequent polyploidisation. In 1971, triticale was separated into individual botanical genus ×*Triticosesale* in familia *Poaceae* (alt. *Gramineae*), subfamilia *Pooideae*, and tribus *Triticeae* with its own species, subspecies, ecological types, varieties, and cultivars (BAUM, 1971; STACE, 1987). Botanical genus ×*Triticosecale* was divided into four groups, depending on the ploidy level, namely tetraploids (2n = 4x = 28 chromosomes), hexaploids (2n = 6x = 42), octoploids (2n = 8x = 56) and decaploids (2n = 10x = 70). Forms with winter, spring, and facultative type of development belong to each group. The most important are three forms of triticale, distinguished by the method of their creation. Two of these forms are recognised as primary bi-species triticale: octoploids (2n = 8x = 56, genome formula AABBDDRR) and hexaploids (2n = 6x = 42, AABBRR). They originated directly from crosses of, respectively, bread wheat (*Triticum aestivum* L., 2n = 6x = 42, AABBDD) or durum wheat (*Triticum durum* L., 2n = 4x = 28, AABB) with rye (*Secale cereale* L., 2n = 2x = 14, RR). A tri-species secondary (or 'hybrid') hexaploid triticale (2n = 42, AABBRR) is distinguished as a separate form. It was originated from crosses of the primary hexaploid triticale with the primary octoploid triticale. Therefore, triticale has synthesised heredity of three species – bread wheat, durum wheat and rye.

The new cereal crop has unified in itself not only positive properties of its parental species, but also some negative traits which triticale breeders have been perseveringly trying to get rid of. The history of triticale creation and improvements amount to nearly 135 years in different countries of the globe. The 'archaic' period of triticale research was finished in 1937 with the discovery of the ability of an alkaloid colchicine to cause chromosome doubling, and with the development of methodology

of chromosome doubling by application of this alkaloid (BLAKESLEE & AVERY, 1937; NEBEL & RUTTLE, 1938). This gave an opportunity to the more-or-less effective creation of amphidiploids in unrestricted amounts.

During the initial period of triticale history, its breeding was mainly focused on octoploid triticale (MÜNTZING, 1936, 1939, 1957, 1963, 1979; PISAREV & VINOGRADOVA, 1944). Long-term studies conducted by A. Müntzing (Institute of Genetics, Lund, Sweden) for the improvement of octoploid triticale resulted in the creation of highly winter hardy, early maturing, and high-protein grain lines with good to excellent bread making qualities, far excelling wheat in adaptation to light soils. However, because of their substantial negative characteristics (low fertility, shrivelled grain, prone to lodging) these octoploid triticale could not compete in grain yield with wheat and rye in relatively favourable conditions. Hopes that were set on octoploid triticale were not justified (MÜNTZING, 1939, 1979; SANCHEZ-MONGE & TJIO, 1954; PISAREV & ZHILKINA, 1967). At the First International symposium of wheat genetics in 1958, the better availability of hexaploid triticale was declared.

The widest programme of triticale breeding was conducted from the end of 1963 in CIMMYT (International Maize and Wheat Improvement Centre, Mexico), initiated by N.E. Borlaug and F.J. Zillinsky as a collaborative research between CIMMYT and University of Manitoba (Canada) (ZILLINSKY & BORLAUG, 1971). The main aim of the breeding was the improvement of triticale in competition with other crops, especially in the betterment of human nutrition in poverty-stricken countries. The collaboration of these establishments have accelerated and stimulated research with triticale around the globe.

In South Africa, R.D. Pienaar initiated triticale breeding during 1960 at the University of Stellenbosch. The breeding programme was based on the initial breeding material from Canada, received from B.C. Jenkins (PIENAAR, 2006). This material was crossed with primary triticale created from local wheat and rye forms. Later this locally created germplasm was used in crosses with triticale lines from France, Hungary, Spain, and USA. In 1963 and 1973, new advanced breeding

material was obtained from B.C. Jenkins. Cultivars BC, Joseph, and Oom Jan were released in 1975. These first cultivars had major disadvantages of being late maturing and tall. The first more successful cultivar Usgen 7 was released in 1979 and was used as forage crop, for hay production and some bread making. However, its yield was too low. After N.E. Borlaug had visited the University of Stellenbosch, the breeding programme obtained a boost in receiving international triticale yield nurseries (ITYN) from CIMMYT in 1975. The material was used in crosses and for direct selections and introductions. In 1985, these selections from the CIMMYT's nurseries were released as cultivars, namely, Usgen 10, Usgen 14, and Usgen 18. This mainly contributed in the observed increase of triticale acreage from 5 000ha to 30 000ha in the Western Cape. During 1990, a selection from the 19th ITYN entry 10 was released as cultivar Usgen 19. In recent years, newer cultivars, such as Ibis, Bacchus, and Tobie were released, which were used for grain, hay, silage production, and as a cover crop in vineyards with the total estimated acreage of 40 000ha. Breeding efforts have been focused on the improvement of yield, grain quality, protein content, and disease resistance of feed cultivars and the selection of lines suitable for grazing, hay, and silage production (ROUX *et al.*, 2006).

The last 20–30 years of triticale breeding were marked with considerable progress, linked with effective recombinant selection based on accumulated genetic polymorphism. High-yielding cultivars were created in Bulgaria (Vihren, Persenk, Mexitol 1, Zaryad), France (Newton, Torpedo, Tropic), Germany (Trimaram, Binova), Romania (Ploi, Colind), Portugal (Crado, Casto, Verdi) and Chile (Calbuco, Antico) (BILITYUK *et al.*, 2004). Worldwide the area under triticale is more than four million hectares, with expected further expansions (MATS'KOVYAK, 1990; GUEDES-PINTO, DARVEY & CARNIDE, 1996; TSVETKOV & STOEVA, 2003). Triticale has acquired its widest expansion in Poland, China, Germany, Australia, Belarus, France, Hungary, Canada, Spain, Russia, and Ukraine. Among all the countries in the world, Poland ranks first in relation to triticale introduction into the agro-industry. During

2005, worldwide a total of 14.7 million tonnes of grain regarding winter and spring triticale combined were harvested (RYABCHOUN *et al.*, 2007).

Modern triticale cultivars are characterised by a unique combination of the best economic-biological properties of its parental species, namely wheat and rye: high potential yield of grain and green biomass; enhanced adaptive properties (higher winter hardiness, ground frost and drought tolerance, unpretentiousness to soils); combined resistance to fungal diseases and pests; high efficiency of nutrient elements usage, higher yield of protein and lysine from an area unit (RYABCHOUN *et al.*, 2007).

Breeding of triticale is carried out in the following directions: grain cultivars for bread making, pastry, fermentation and mixed fodder industries; grain-hay cultivars for green biomass, forage, mixed fodder for cattle, poultry and fish; hay-harvest cultivars exclusively for green fodder, pasture, hay, silage, haylage. Bread products made from triticale flour occupy a special place in child and dietary nutrition. Triticale flour in a mixture with low quality wheat flour – in proportion of 1:5 or 1:4, up to 1:1 – is used as an improver of bread quality (RYABCHOUN *et al.*, 2007).

Triticale grain is widely used for the production of ethanol, which is used for medical, potable, and technical applications. From triticale grain harvested from 100 000ha annually it is possible to produce 210–240 thousand tonnes of ethanol (RYABCHOUN *et al.*, 2007). Triticale grain is characterised by better physicochemical properties for ethanol production compared to wheat. High self-saccharification and liquefaction of triticale mash results in more complete fermentation of carbohydrates and a higher ethanol yield compared to wheat (Table 2.1.1; after FARADZHEVA, 2000; RYABCHOUN *et al.*, 2007). The ethanol yield from triticale grain is 1.66–1.90% higher than from rye and 0.33–0.57% higher than from wheat (RYABCHOUN *et al.*, 2006). According to the Ministry of Agrarian Policy of the Ukraine, only maize is superior to triticale in ethanol yield from a tonne of grain. However, because of the fact that the average triticale yield in the country is higher, the cost of its production per unit area is also considerably lower, which makes triticale the most effective crop for the

production of ethanol. It is also worthwhile to consider the higher nutritional value of by-products (DDGS) as animal feedstock compared to by-products from other crops. Thus, the introduction of triticale as a more effective and ecologically clean feedstock for the ethanol production could help to decrease costs and improve the ethanol yield and quality.

Table 2.1.1. Comparative characteristics of mature wort from different crops (after FARADZHEVA, 2000; RYABCHOUN *et al.*, 2007)

Wort characteristics	Crop, cultivar		
	Triticale, Talva 100	**Wheat, Tarasovskaya 29**	**Rye, Talovskaya 15**
Carbohydrates, %			
Soluble	0.26	0.29	0.30
Insoluble	0.05	0.07	0.07
Loses of fermentable carbohydrates, % of starch			
Insoluble carbohydrates	2.27	2.74	2.68
Insoluble starch	0.46	0.56	0.54
Ethanol output, L.tonne^{-1} of 'conditional' starch	667	663	662

Spring triticale is better adapted crop to the growth conditions than spring wheat, and can be successfully propagated after grain and silage maize, soya bean (*Glycine max*), sugar beetroot and other hoed crops, such as sunflower (*Helianthus annuus*). When spring triticale is planted after a stubble rotation the losses of grain yield to pests and diseases are minimised compared to winter wheat. Spring triticale is effectively used as an insurance crop when the re-sowing of winter cereals is necessary due to winter kill (RYABCHOUN *et al.*, 2007). Triticale is deservedly recognised as the most adapted crop for making the plant production environmentally 'greener.' Its high yield potential, low resource and energy intensity, high earning capacity, and short period of cost turnover expands the opportunities for clean agricultural production and the stabilisation of ecologically safe food grain market.

Nowadays it is not too difficult to talk about triticale and its future as a crop. Competitive cultivars of the new crop do exist and are being steadily improved. During the last decades, triticale has become one of the most promising, high yielding

cereal crops, with its global acreage steadily increasing. In breeding laboratories of the world the search for effective methods of new initial material creation, technologies of propagation and triticale grain processing for various industrial sectors are intensified.

2.2. CONTENT AND STRUCTURAL ORGANISATION OF THE CARBOHYDRATE COMPLEX IN CEREAL GRAIN

> "There are A, B and C glucan chains in amylopectin, A and B polymorphs in granules, A and B granules in some species and there are A, B and C types of starch" (WANG, BOGRACHEVA & HEDLEY, 1998)

2.2.1. General grain composition

Average cereal grain is composed of 12–14% water, 7–12% protein, 65–75% carbohydrates, and 2–6% lipids, which means that they are low in protein and high in carbohydrates (HAARD *et al.*, 1999). Cereal endosperm is represented by starch-filled interior tissues and an external epidermal layer (aleurone), where most of the grain phosphate is located in the form of phytin particles in protein bodies (FALK *et al.*, 2001). Among the storage compounds of cereal grain, starch is quantitatively predominant, with free sugars also present but in small quantities (about 2–3% of dry matter in wheat grain; LINEBACK & RASPER, 1998). Waxy (amylose-free) cultivars of barley are characterised by considerably higher levels of maltose because of higher levels of α-amylase enzyme (XU *et al.*, 1997). Starch production is critical to both the yield and the quality of the grain. Starch content in the grain varies depending on genotype and the environmental conditions in a wide range (TYMCHOUK *et al.*, 2002). In maize, for instance, it can range from 38 up to 72% (SHMARAEV, 1975), which creates opportunities for targeting this trait for improvement through breeding and selection. As a rule, in favourable growing conditions the starch content in the grain increases, and is positively correlated with grain yield (SOZINOV & ZHEMELA, 1983). In studies with wheat, soil water deficit decreases the contents of both starch and amylose in grain, but increases the protein content (DAI *et al.*, 2008).

2.2.2. Starch composition

From its chemical structure, starch is a typical polymer and consists of 96.1–97.6% polysaccharides, which, if acidically hydrolysed, creates glucose (GUILBOT & MERCIER, 1985). On the polysaccharide matrix of the starch granules, some non-starch components, namely lipids, proteins and mineral substances are also absorbed. Lipids are represented by free fatty acids such as palmitic, linoleic and stearic, and lysophospholipids, which are associated with amylose and are mainly located at the surface of starch granules (BALDWIN, MELIA & DAVIES, 1997). Percentage wise, lipids are the most predominant non-starch component in the starch granule. Their content ranges from less than 0.001 up to 0.6% and even 1.5%, but normally within the range of 0.005–0.010% (GUILBOT & MERCIER, 1985; ELLIS *et al.*, 1998; BULEON *et al.*, 1998; HOOVER & VASANTHAN, 1994; VASANTHAN & BHATTY, 1996; SAHLSTRÖM *et al.*, 1998; ANDERSSON *et al.*, 1999b; ABDEL-AAL *et al.*, 2002; TESTER, KARKALAS & QI, 2004a). Larger starch granules have lower lipid contents than smaller ones (RAEKER *et al.*, 1998). Starch-associated lipid complexes may reduce starch digestion by enzymes (VASANTHAN & BHATTY, 1996). Waxy starches are known to possess lower levels of phospholipids (YASUI *et al.*, 1996; HAYAKAWA *et al.*, 1997; ABDEL-AAL *et al.*, 2002; GEERA *et al.*, 2006; SAHLSTRÖM, BÆVRE & GRAYBOSCH, 2006).

Starch granules contain up to 0.003% protein (CORNELL *et al.*, 1994; HOOVER & VASANTHAN, 1994; VASANTHAN & BHATTY, 1996; ABDEL-AAL *et al.*, 2002), the content of which is also shown to be higher at the surface of the granule (BALDWIN, 2001). In wheat, friabilin proteins (e.g. puroindolin isoforms PIN-a and PIN-b) affect endosperm hardness through bonding properties of starch granules to matrix protein. Puroindolins are more abundant in soft than in hard wheat, and absent in *Triticum durum* (GREENWELL & SCHOFIELD, 1989; MORRIS *et al.*, 1994; ODA & SCHOFIELD, 1997; BALDWIN, 2001). Absence of both puroindolin isoforms, or presence of PIN-a combined with mutations of PIN-b leads to endosperm hardness (SMITH *et al.*, 2006). It was also found that in wheat two starch granule-bound proteins (SGP-140 and

SGP-145) were mainly associated with A-type starch granules (>10μm). Such tendency was confirmed for other cereals like rye, barley and triticale (PENG *et al.*, 2000).

Starch contains up to 0.2–0.7% mineral substances, which are represented mainly by phosphoric acid (with the highest content in starch of potato, *Solanum tuberosum*) linked by a complex ester with the amorphous carbohydrate part (GUILBOT & MERCIER, 1985; LORBERTH *et al.*, 1998; BLENNOW *et al.*, 2000, 2002). Starches from cereals contain negligible amounts of mineral fraction (SVIHUS, UHLEN & HARSTAD, 2005). Lower ash content was found in starch of waxy hexaploid wheat compared to wild-type wheat (ABDEL-AAL *et al.*, 2002).

2.2.2.1. Starch complex structure and composition in details

Starch is a polymorphic polyglycoside, which consists of two co-polymers – amylose and amylopectin. They differ in the level of their polymerisation, positions of chemical links that unite the monomers, and their basic physicochemical and technological properties. In cereals roughly three-quarters of the total starch is amylopectin (Figures 2.2.2.1.1 and 2.2.2.1.2; CHAPLIN, 2007), which consists of branched glucose chains that form insoluble, semi-crystallic granules (BEMILLER & WHISTLER, 1996). The remainder of the starch molecule consist of amylose (Figure 2.2.2.1.3; CHAPLIN, 2007), which is composed of linear chains of glucose that adopt a double or single helical configuration within the granule (BULEON *et al.*, 1998; MYERS *et al.*, 2000). In addition, there is also a small proportion of an intermediate fraction, which consists of branched amylose molecules and small amylopectin molecules (RAHMAN *et al.*, 2007).

Figure 2.2.2.1.1. Representative partial structure of amylopectin (CHAPLIN, 2007)

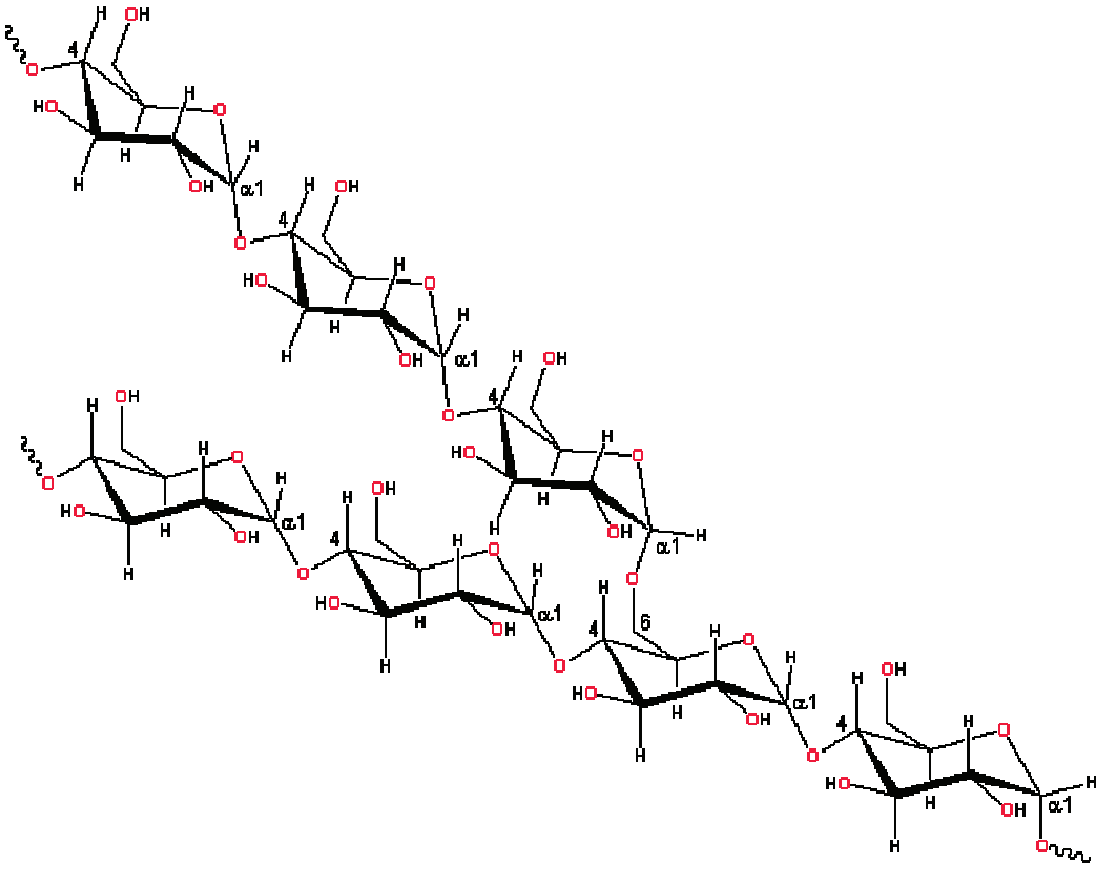


Figure 2.2.2.1.2. Amylopectin model structure (CHAPLIN, 2007)

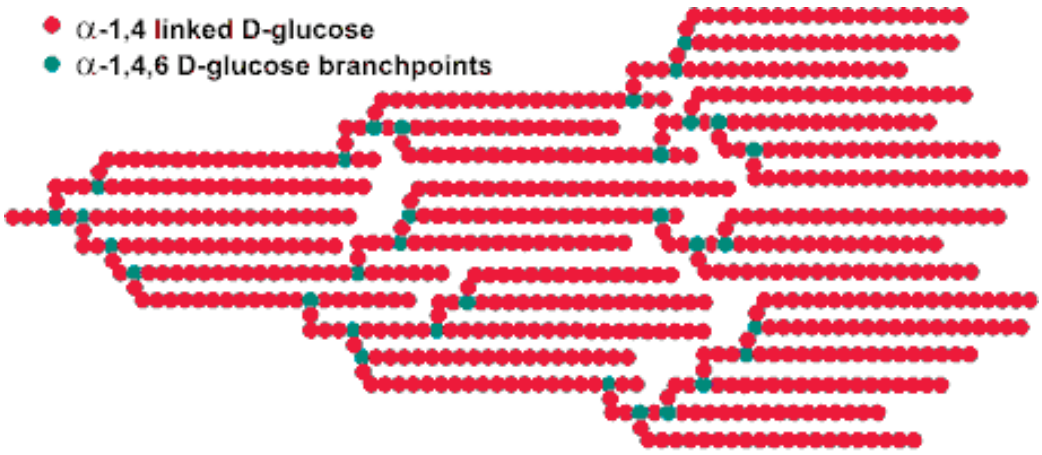
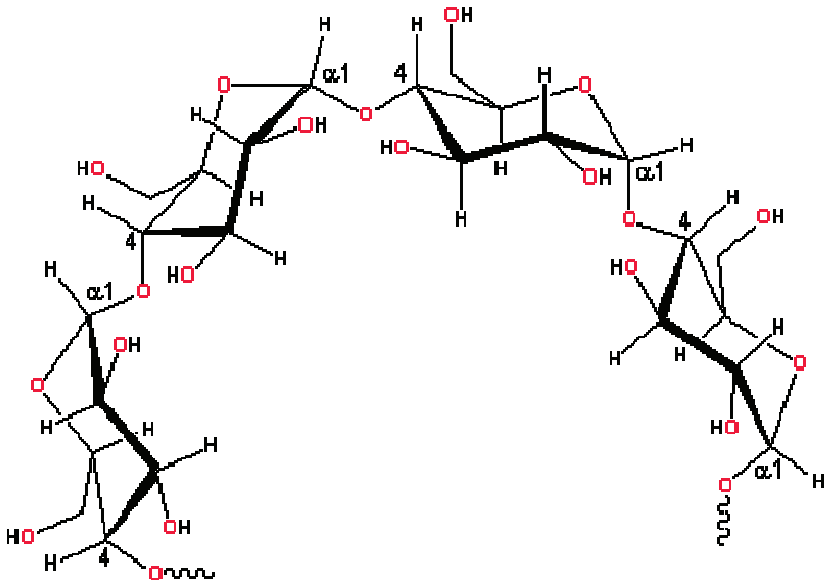


Figure 2.2.2.1.3. Representative partial structure of amylose (CHAPLIN, 2007)



Amylose content has substantial genetic variability and naturally ranges from 0% to about 40–50% in major cereals such as barley (MORRISON, SCOTT & KARKALAS, 1986; BJORCK *et al.*, 1990; SALOMONSSON & SUNDBERG, 1994; VASANTHAN & BHATTY, 1996; BHATTY & ROSSNAGEL, 1997; AKERBERG, LILJEBERG & BJORCK, 1998; ANDERSSON *et al.*, 1999a; MOHAMMADKHANI, 2005), rice (*Oryza sativa* L.; SANO, KATSUMATA & OKUNO, 1986; NAKAMURA *et al.*, 1995), rye (MOHAMMADKHANI, STODDARD & MARSHALL, 1999a), wheat (NAKAMURA *et al.*, 1995; KIRIBUCHI-OTOBE *et al.*, 1997; YASUI *et al.*, 1997; JANE *et al.*, 1999; YAMAMORI *et al.*, 2000; MOHAMMADKHANI, STODDARD & MARSHALL, 1998; PENG *et al.*, 1999; ABDEL-AAL *et al.*, 2002) and its wild relatives (MOHAMMADKHANI, STODDARD & MARSHALL, 1999a; RODRIGUEZ-QUIJANO, VAZQUEZ & CARRILLO, 2004). In the unmodified wild-type maize starch amylose content is about 22–27%, and amylopectin respectively 73–78% (GUILBOT & MERCIER, 1985), but can reach up to 90% in some mutant forms (FERGASON, 1994; SIDEBOTTOM *et al.*, 1998; PARKER & RING, 2001). Starches of other cereal crops have approximately the same fractional composition, in contrast with the leguminous starches that are characterised by increased amylose content of up to 30–32% (HEDLEY *et al.*, 1996). Average amylose

content in starch of a different origin is described in Table 2.2.2.1.1 (FAO, 1998) and Table 2.2.2.1.2 (POWER, 2003).

Table 2.2.2.1.1. Some properties of whole granular starches (FAO, 1998)

Source	Gelatinisation temperature range, °C	Granule shape	Granule size (mm)	Iodine binding capacity (g I_2/100g)	Amylose content (%)
Barley	51–60	Round or lenticular	20–25, 2–6	4.3	22
Triticale	55–62	Round	19 (2–35)	-	23–24
Wheat	58–64	Lenticular or Round	20–35, 2–10	5.0	26 (23–27)
Rye	57–70	Round or lenticular	28 (12–40)	5.5	27
Oats	53–59	Polyhedral	5–10	5.1	23–24
Potato	59–68	Oval	40 (15–100)	4.5	23
Maize	62–72	Round or polyhedral	15 (5–25)	5.3	28
Waxy maize	63–72	Round	15 (5–25)	0.1	1
Broad bean	64–67	Oval	30	4.5	24
Sorghum	68–78	Round	15–35	-	25 (23–28)
Rice	68–78	Polygonal	3–8	-	17–19* 21–22**
High amylose maize	67–80	Round or Irregular sausage shaped	25	*ca*. 10.5	52
Peas smooth	55–70	Reniform*** (simple)	5–10	6.7	33–36
Peas wrinkled	>99	Reniform*** (compound)	30–40	14.7	71–76

* japonica; ** indica; *** kidney-shaped.

Table 2.2.2.1.2. Typical percentage of amylose and amylopectin in starches from different crops (POWER, 2003)

Starch source	Amylose, %	Amylopectin, %
Wheat	25	75
Potato	20	80
Tapioca/cassava/manioc	17	83
Rice	20	80
Waxy rice	2	98
Maize	25	75
Waxy maize	1	99
High amylose maize	50-75	25-50
Sorghum	25	75
Waxy sorghum	<1	>99
Heterowaxy sorghum	<20	>80

Starch structures are also influenced by growth temperature, which may change the amylose/amylopectin ratio, the molecular structure of these polymers, and distributions of the amylopectin chain length (GERNAT *et al.*, 1993; TESTER & KARKALAS, 2001; KOHYAMA & SASAKI, 2006). Higher temperatures during cereal grain filling result in reduced starch synthesis (lesser starch content in endosperm, smaller starch granules), a higher amylose content and a higher gelatinisation temperature, as was reported for wheat (TESTER *et al.*, 1991; TESTER *et al.*, 1995). For barley, the influence of the growth temperature on amylose content is ambiguous (TESTER & KARKALAS, 2001), and for maize and rice it was reported that amylose content in starch decreases as average growth temperature rises (FERGASON & ZUBER, 1962; ASAOKA *et al.*, 1984; SANO, MAEKAWA & KIKUCHI, 1985; UMEMOTO, NAKAMURA & ISHIKURA, 1995; YANAGISAWA, KIRIBUCHI-OTOBE & FUJITA, 2004). However, another report for rice showed a high positive correlation between average maximum air temperature and amylose content in rice types with high apparent amylose contents (AAC), and negative correlation in types of rice with low and intermediate AAC (CHEN *et al.*, 2008).

2.2.2.2. Amylose and amylopectin structures

Amylose is a long, essentially linear polymer of glucose with occasional branching points, in which D-glucopyranosyl monomers are linked between each other by α-1,4 glycoside links, with a degree of polymerisation (DP; also called average chain length) in the range of 500–6000 glucose residues; a small fraction of the amylose molecules is slightly branched by α-1,6 glycoside links (HIZUKURI, TAKEDA & YASUDA, 1981; TAKEDA & HIZUKURI, 1987; SHIBANUMA *et al.*, 1994; WANG, BOGRACHEVA & HEDLEY, 1998). Amylose of wheat has a DP of about 300 residues with an average 1.9 branches per molecule (TAKEDA, SHIRASAKA & HIZUKURI, 1984).

Amylopectin, on the other hand, is an extensively branched glucan with a DP ranging from 3×10^5 to 3×10^6 glucose monomers (ZOBEL, 1988a), with branch chains

of different lengths (HIZUKURI, 1986). In these branch chains after every 15–45 monomers of glucose non-random α-1,6 glycoside links are present. These links connect the linear amylose-type α-1,4 D-glucose chains with each other (MANNERS, 1989; RAHMAN *et al.*, 2007). Branches are clustered and occur about every 9nm (BULEON *et al.*, 1998). For wheat and rice the average branch length is 11nm (O'SHEA *et al.*, 1998; UMEMOTO *et al.*, 2002) and for maize 13nm (PERERA *et al.*, 2001; RAHMAN *et al.*, 2007). At the expense of spatial interaction of neighbouring chains amylopectin molecule acquires not only branching, but also a spiralled (helical) structure (THOMPSON, 2000).

Amylopectin molecules contain about 10 times more glucose residues than amylose. Relative molecular mass of amylose is approximately 10^4–10^5 units, when amylopectin relative molecular mass is about 10^7–10^8 units (GIDLEY & BOCIEK, 1985). Amylopectin from waxy starch has a larger weight-average molecular weight than amylopectin of normal starch (YOO & JANE, 2002b). Amylopectin is one of the biggest natural polymers and by molecular weight rebate probably only to glycogen (JENKINS *et al.*, 1993). On average each amylopectin molecule contains up to two million glucose residues, forming a compact structure with a radius of 21–75nm (PARKER & RING, 2001); of these residues about 5% form the branching points with α-1,6 linkages (JOBLING, 2004; TESTER, KARKALAS & QI, 2004a).

2.2.3. Non-starch polysaccharides

In addition to starch, cereal endosperm contains considerable amounts of non-starch polysaccharides (NSP), which are deposited in the endosperm cell walls (RUDI *et al.*, 2006). 80% of the whole grain NSP are found in its bran (SMITH *et al.*, 2006). Carbohydrate complexes of oats, barley, and rye contain a relatively high proportion of NSP (5–25% of total carbohydrates), which consists of pentosans of different molecular weight (WOODS, WEISZ & MAHN, 1991). Cereal pentosan fraction is a complex mixture of fructan and β-glucan (BG) with arabinoxylan as main ingredient; it also contains small amounts of glucose and ferulic acid (HAARD *et al.*, 1999; SMITH

et al., 2006). BG in cereals is a linear mixed-linkage glucan which consists mainly of cellotriose and cellotetraose linked by β-1,4 linkages, with these blocks being interlinked by occasional β-1,3 linkages (BUCKERIDGE *et al.*, 2004; RUDI *et al.*, 2006). Wheat contains lesser amounts of pentosans, and has a higher xylose/arabinose ratio than rye (ELIASSON & LARSSON, 1993). Total NSP content in the whole grain wheat is 11%, arabinoxylans being the largest part of it representing 8% (DWB) of the grain weight (ENGLYST *et al.*, 1992). Arabinoxylans represent 70% of the wheat endosperm cell walls (SORENSEN, PEDERSEN & MEYER, 2006). Quantitative trait loci (QTL) were identified for water extractable arabinoxylans in wheat (MARTINANT *et al.*, 1998, 1999). In research done with New Zealand' wheat, it was shown that the starch content of grain is negatively correlated to arabinoxylan content (COLES *et al.*, 1997). Waxy wheat was found to possess increased levels of the substances arabinoxylan and polyphenol (TAKATA *et al.*, 2005, 2007). Double addition of rye chromosomes 2R and 5R into the wheat genome leads to the increase of soluble dietary fibre (SDF) content and soluble non-cellulosic glucose in the NSP fraction above the rye level, while lines with the addition of 3RS chromosome arm show decreased SDF content below the wheat level (CYRAN, RAKOWSKA & MIAZGA, 1996).

While arabinoxylans alone are the major NSP component of wheat and rye cell walls, arabinoxylans and BG are the dominant components in barley and oats (HOLTEKJØLEN *et al.*, 2006). BG content has a strong genetic influence, although considerable effects of environmental factors on it are also found (BRUNNER & FREED, 1994; PEREZ-VENDRELL *et al.*, 1996; ZHANG *et al.*, 2001). In barley, the variation in BG content was found to extend from 3 to 20% (HOLTEKJØLEN *et al.*, 2006; RUDI *et al.*, 2006). In barley mutants with either the high-amylose or the high-amylopectin endosperm, the BG content is significantly higher in comparison to cultivars with conventional amylose/amylopectin ratios (OSCARSSON *et al.*, 1996; XU *et al.*, 1997; RUDI *et al.*, 2006). In addition, it was shown that in the high lysine barley mutants with low total starch content (29.8%) BG has compensating effects (BG content rises

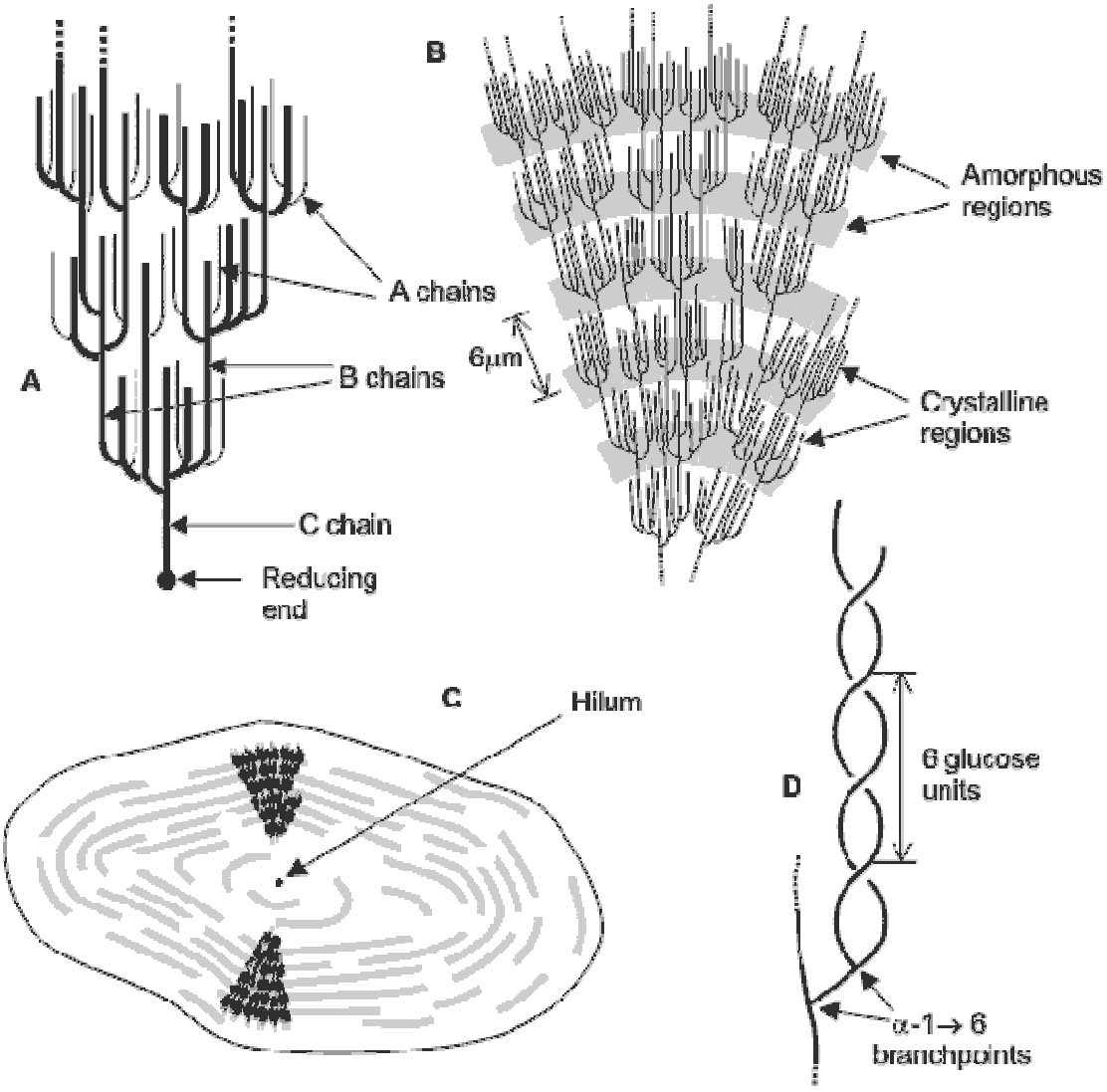
to 19.8%) which gives a sum of total starch and BG content as high as 49.4% (MUNCK *et al.*, 2004).

2.3. STRUCTURE OF STARCH GRANULES OF CEREAL ENDOSPERM AND ITS DEPENDENCE ON STARCH FRACTIONAL COMPOSITION

Starch is deposited in plant cells not in homogenous form, but as semi-crystalline starch granules (Figure 2.3.1; CHAPLIN, 2007), and their microstructure was subject of special research (BULEON *et al.*, 1998; THOMPSON, 2000; DONALD, 2001). Starch granules in polarised light behave as optically positive spheroids and show a picture that is characteristic for objects with rod-like architecture. This evidences that they are composed of radial chains of glycosides (FRENCH, 1984). This interpretation of molecular structure of starch was confirmed by an X-ray diffraction analysis. The X-ray structural analysis also showed that in amylose molecule a number of parallel polyglycoside chains are interlinked, some of which have spiral structures (DENYER *et al.*, 2001). Because of this, amylose fraction of starch consists of linear and branched molecules (TAKEDA *et al.*, 1987; BILIADERIS, 1991). Ratio of linear and branched molecules can vary. Starches of cereal crops have a lower ratio than starches of tuberous plants; for example, maize starch has a ratio of 1:5.3, and cassava (*Manihot* spp.) starch has 1:17.1 ratio (TAKEDA *et al.*, 1993).

It is evident that formation of starch granules in endosperm is a direct result of starch structural co-polymers synthesis (FRENCH, 1984; SMITH *et al.*, 1997), and starch characteristics are related to the chemical structures of the amylopectin and amylose and the way in which they are arranged in the starch granule. Starch granule synthesis starts from the formation of a little initial quantity of starch (granule nuclei) in the stroma of amyloplast, which consists of numerous tubes and tylacoids (BADENHUIZEN, 1965; SHANNON, CREECH & LOERCH, 1970). In some cases, tubes and tylacoids surround such a starch zone, which is located in the centre of amyloplast, and form around it an open or closed circle. As a result a structure is formed which looks like a vacuole (BADENHUIZEN, 1963).

Figure 2.3.1. Starch granule (CHAPLIN, 2007)



In the figure above:
A: The essential features of amylopectin.
B: The organisation of the amorphous and crystalline regions (or domains) of the structure generating the concentric layers that contribute to the 'growth rings' that are visible by light microscopy.
C: The orientation of the amylopectin molecules in a cross section of an idealised entire granule.
D: The likely double helix structure taken up by neighbouring chains and giving rise to the extensive degree of crystallinity in granule.

Newly synthesised starch is deposited in stroma and surrounds the initial starch grain, which results in the creation of the centre of starch formation, which is called a hilum. Two main types represent starch granules, namely: simple granules and compound granules. Simple granules have one centre of emergence (i.e. hilum) and in most cases are typical for potato, maize, and wheat. On the other hand, compound granules emerge from more than one hilum and are typical for oats and

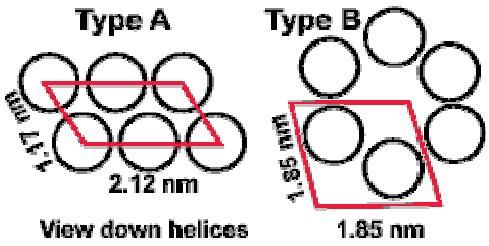
rice (TKACHENKO & SERBIN, 1997; SERBIN *et al.*, 2003). A hilum quickly grows by apposition (YOSHIDA *et al.*, 1958; DENYER *et al.*, 1996), and finally all the amyloplast fills up with starch. During the process amylose is synthesised side by side with amylopectin deep inside the granule (JANE, 2006), and the granule itself becomes bigger because of amylopectin synthesis in the direction of the granule surface (BABA, YOSHII, & KAINUMA, 1987; DENYER *et al.*, 2001). The tubes, which are located in stroma, are pushed aside towards the plastid circumference. Starch grain keeps growing; its membrane stretches and eventually becomes a dry film, which can act as a physical barrier to enzymatic digestion (FISHER *et al.*, 1997; SVIHUS, UHLEN & HARSTAD, 2005).

Starches of cereals have amylose of smaller molecular sizes than starches from tubers and roots (TAKEDA & HIZUKURI, 1987; JANE & CHEN, 1992). Amylose composes an internal amorphous phase of starch granule (KAIZUMA, 1988; KUIPERS *et al.*, 1994). However, a little amount of amylose is also present in its semi-crystalline area (JENKINS & DONALD, 1994). Amylose molecules are cross-linked with amylopectin (JANE *et al.*, 1992). By application of partial gelatinisation it was shown that amylose of the external part of starch granule is more concentrated, and has a lower molecular weight than amylose from internal parts of the granule (JANE & SHEN, 1993). Branch chains of amylopectin at the circumference of a starch granule are also shorter than in the interior part of the granule (JANE *et al.*, 2006). It was also demonstrated that in starch of potato and maize, individual amylose molecules are scattered among amylopectin molecules (JANE *et al.*, 1992). With the maturation of seeds the amylose content of starch and the size of starch granules increases (MORRISON & GADAN, 1987; JANE & SHEN, 1993; PAN & JANE, 2000; YOSHIDA *et al.*, 2003). Thus, it was concluded that amorphous amylose molecules are interspersed amongst structured semi-crystalline amylopectin and are more concentrated at the circumference of a starch granule than at its core (JANE *et al.*, 2006). It results in higher concentration of amylose at the periphery of starch granules. There amylose and amylopectin molecules interact more intensively, which contributes to a lesser

susceptibility of uncooked normal starch granules to enzymatic hydrolysis and their slower digestibility. It is also known that high-amylopectin starches are more easily digestible than normal or high-amylose starches (XUE *et al.*, 1996; AKERBERG *et al.*, 1998; ANKRAH *et al.*, 1999; ITO *et al.*, 1999; BEDNAR *et al.*, 2001; SAITO *et al.*, 2001; ABDEL-AAL *et al.*, 2002; JANE *et al.*, 2003; VIGNAUX *et al.*, 2004). Reduced enzymatic digestibility of high-amylose starches could be explained by a formation of an amylose-lipid complex on the surface of granules (CUI & OATES, 1999; CROWE, SELIGMAN & COPELAND, 2000; TUFVESSON *et al.*, 2001).

Clusters of parallel amylopectin chains, which are twisted in left-handed double spirals (helices) of neighbouring molecules, with six glucose residues per turn, are able to stack next to each other which leads to the creation of a semi-crystallic (crystalline) area in a starch granule (IMBERTY *et al.*, 1988; IMBERTY & PEREZ, 1988; IMBERTY *et al.*, 1991; BULEON *et al.*, 1998; VERMEYLEN *et al.*, 2004; KISELEVA *et al.*, 2005; BERTOFT, 2007a, 2007b; KOZLOV *et al.*, 2007a). Wild-type starch granules have 15–45% crystallinity (ZOBEL, 1988b). These double-helical chains can form three distinct types of polymorphic crystallites (A, B and C), which are characteristic to certain plants, depending on their amylopectin branch chain length and packing structure, which was revealed by an X-ray diffraction picture of starch grains (HIZUKURI, 1985; IMBERTY *et al.*, 1991; GALLANT, BOUCHET & BALDWIN, 1997; WANG *et al.*, 1998; KUBO *et al.*, 2008). The A-type crystallites have a denser, staggered monoclinic packing, with a minimal amount of bound water (Figure 2.3.2; CHAPLIN, 2007).

Figure 2.3.2. Amylopectin of A and B types (CHAPLIN, 2007)



These A-type crystallites have amylopectin with unbroken chain lengths of about 23–29 glucose units. Most cereals (e.g. maize, rice, and wheat) belong to the A-type starches. B-type crystallites have a more open hydrated hexagonal packing. They have longer unbroken chain lengths of amylopectin (approximately 30–44 glucose residues), which extends through two, three or more clusters (i.e. has B2, B3 and longer chains) (HIZUKURI, 1986). B-type crystallites are found in high-amylose cereal starches, banana (*Musa* spp.), tuberous and bulbous plants (e.g. potato, *Canna* spp., *Lilium* spp.) (ZOBEL, 1988a; IMBERTY *et al.*, 1991; BILIADERIS, 1991; GERNAT *et al.*, 1993; TANG, MITSUNAGA & KAWAMURA, 2006). C-type crystalline structure is a combination of A and B types which is found in beans (*Phaseolus* spp.), peas (*Pisum* spp.) and sweet potato (*Ipomoea batatas*) (CAIRNS *et al.*, 1996; WANG *et al.*, 1998; TANG, MITSUNAGA & KAWAMURA, 2006; KUBO *et al.*, 2008). Some amylopectins are characterised by an alternative arrangement of interconnected clusters (BERTOFT, 2004).

Starches with different types of polymorphism show different levels of enzymatic digestibility. The A-type polymorphic starch is easily digestible, in contrast with the B-type and some of C-type starches which are very resistant to enzymatic hydrolysis (FUWA, TAKAYA & SUGIMOTO, 1980; KIMURA & ROBYT, 1995; SPENCE & JANE, 1999; PERERA *et al.*, 2001; JANE *et al.*, 2003). B-type starches are also known to be more resistant to pressure than A or C type starches (STUTE *et al.*, 1996; KATOPO, SONG & JANE, 2002; OH *et al.*, 2008).

However, such crystalline structure is not a property of an entire starch granule. Amylopectin molecules are located radially inside of starch granules, and thus their ends are oriented towards the surface of granules (FRENCH, 1984). Because of their radial orientation, and because the radius of the granule increases with maturation, the number of amylopectin branches required to fill up the spaces between molecules also increases. This results in the formation of concentric layers (120–500nm thick) of alternated amorphous (amylose and amylopectin molecules in disordered conformation) and semi-crystalline (alternated amorphous and crystalline

lamellae, with repeat distance of about 9nm) growth rings inside the granule (DENYER *et al.*, 2001; PILLING & SMITH, 2003; VANDEPUTTE & DELCOUR, 2004; YURYEV *et al.*, 2004; VERMEYLEN *et al.*, 2006; KOZLOV *et al.*, 2007a, 2007b; KOROTEEVA *et al.*, 2007). It was suggested that these growth rings are laid down at a rate of one layer per day due to variation in photosynthetic activity and as the result differential access to glucose (TESTER, 1997b; SMITH, 2001).

The crystalline and amorphous lamellae of semi-crystalline amylopectin are organised into 'blocklets' – larger, more or less spherical structures (GALLANT, BOUCHET & BALDWIN, 1997). Waxy starch granules were shown to have a higher ratio of crystalline to amorphous regions (HAYAKAWA *et al.*, 1997; ABDEL-AAL *et al.*, 2002; KIM *et al.*, 2003). There are normally slightly more 'outer' unbranched chains of amylopectin (named A-chains) than 'inner' branched chains (named B-chains). There is only one chain (called the C-chain) which contains the single reducing group, which begins in hilum, the centre of a starch granule (CHAPLIN, 2007).

Starch granules size, as well as its fraction composition, has a great importance on ensuring a starchy raw material quality (KNUTSON *et al.*, 1982; OKECHUKWU & RAO, 1995; SAHLSTRÖM *et al.*, 1998; PARK, CHUNG & SEIB, 2004, 2005; GEERA *et al.*, 2005b, 2006). Starch granules size distribution varies between cereal crop cultivars and mainly depends on genetically determined starch content in seeds and the percentage of amylose in the starch (SAHLSTRÖM *et al.*, 1998; PETERSON & FULCHNER, 2001). Cereal starch possesses two distinct starch granule groups, namely A and B types with different granule sizes, molecular structure (JANE *et al.*, 2003), compositions (RAEKER *et al.*, 1998; LU, PAI & LU, 1999; PENG *et al.*, 1999; SHINDE *et al.*, 2003), and properties (SEIB, 1994; RAEKER *et al.*, 1998; HAYASHI *et al.*, 2004; HUNG & MORITA, 2005). In wheat, rye, triticale and barley starch granules have typical bimodal distribution with large A-type lenticular granules of 15–45µm in diameter and the smaller B-type spherical fraction of 1–10µm. Although, some researchers divide starch granule size distribution into three groups; the smallest in size and number (up to 3% of the total starch weight) among these is the C-type starch

group, which is actually considered as a sub-group of B-type starch (BECHTEL *et al.*, 1990; LIM *et al.*, 1992; JANE *et al.*, 1992, 1994; VASANTHAN & BHATTY, 1996; PENG *et al.*, 1999; ANDERSSON *et al.*, 1999a; TAKEDA *et al.*, 1999; TANG *et al.*, 2000, 2001; SONG & JANE, 2000; STEVNEBØ, SAHLSTRÖM & SVIHUS, 2006).

In developing cereal endosperm, only one A-type starch granule is produced in each amyloplast (A-type amyloplast) during 4^{th} to 14^{th} days after anthesis, when the endosperm cells are still dividing and sufficient quantities of enzymes are available for rapid synthesis of glucans (BRIARTY, HUGHES & EVERS, 1979; BECHTEL *et al.*, 1990; PENG *et al.*, 1999; NOWOTNA *et al.*, 2007). B-type starch granules are synthesised in the protrusions (named stromules) which extend from A-type amyloplast after two weeks past anthesis to maturity, during the endosperm cell expansion stage (PARKER, 1985; PENG *et al.*, 2000; LANGEVELD *et al.*, 2000; BECHTEL & WILSON, 2003; TETLOW, 2006). The larger (>9.8μm), A-type starch granules compose on average up to 3% of the total number of starch granules in wheat endosperm, which amounts to up to 50–87% of the total endosperm starch in weight. The smaller in size (<9.8μm) B-type starch granules, on the other hand, account for up to 99% of the total starch granules number, but contribute to only about 25–30% of the total weight of mature endosperm starch (EVERS & LINDLEY, 1977; BECHTEL *et al.*, 1990; RAEKER *et al.*, 1998; PENG *et al.*, 1999; STODDARD, 1999; SHINDE, NELSON & HUBER, 2003; GEERA *et al.*, 2005a; DAI *et al.*, 2008).

Disc or lenticular shaped A-type starch granules of triticale, wheat and barley consist of higher number of B2 amylopectin chains (which extend through two clusters; they are characterised by a cylindrical-shaped molecules which fit better to the disc-shaped granule) and fewer short A and B1 chains (TAKEDA *et al.*, 1999; JANE *et al.*, 1999; AO & JANE, 2007). These A-type granules are easily hydrolysed by amylases because they have loosely packed internal structures, in contrast to smaller B-type granules (JANE *et al.*, 2003). On the contrary, spherical B-type starch granules have more cone-shaped short A and B1 chains and fewer B2 chains of amylopectin, and have solid internal structures which are resistant to enzyme hydrolysis (TANG,

WATANABE & MITSUNAGA, 2002; JANE, 2006). The B-type starch granules show lower (usually 2–3% less) amylose content and higher level of phospholipids and granule-associated proteins compared to A-type granules (MEREDITH, 1981; MORRISON & GADAN, 1987; RAEKER *et al.*, 1998; SAHLSTRÖM *et al.*, 1998; LU, PAI & LU, 1999; PENG *et al.*, 1999; GAINES, RAEKER & TILLEY, 2000; SHINDE *et al.*, 2003; GEERA *et al.*, 2006; DAI *et al.*, 2008). It was shown that higher level of nitrogen fertilisation leads to increased percentage of B-type granules in starch (NOWOTNA *et al.*, 2007). Large A-type starch granules are more predominant in soft wheat than hard wheat (RAEKER *et al.*, 1998; CAPOUCHOVA & MARESOVA, 2003). It was found that the total starch content (sum of A and B type granules) is more important for ethanol output than their relative amounts (BROSNAN *et al.*, 1999).

Starch granules are found to have holes (pores, channels with diameter from 70 to 300nm) on their surface, scattered in a random order (FANNON, HUBER & BEMILLER, 1992, 1993; BALDWIN *et al.*, 1994; BALDWIN, DAVIES & MELIA, 1997; BALDWIN *et al.*, 1998; JUSZCZAK, FORTUNA & KROK, 2003). The A-type starch granules have pinholes on the surface and serpentine-shaped channels inside of the granule, but the B-type granules do not demonstrate these characteristics (FANNON, HUBER & BEMILLER, 1992; GALLANT, BOUCHET & BALDWIN, 1997). Confocal laser-scattering microscopy revealed that sorghum (*Sorghum* spp.) and maize starch display a large number of such channels (HUBER & BEMILLER, 1997, 2000; GRAY & BEMILLER, 2004). Such pores or pinholes were also confirmed on triticale starch granules by means of atomic force microscopy (NC-AFM) (JUSZCZAK, 2003). These pinholes and channels are most probably produced because of digestion by a native amylase and hydrolysis of the starch granules surface during seed maturation (LI, 2006; JANE, 2006). A higher starch content and percentage of amylose in it leads to a smaller size of the starch granules (MORRISON, SCOTT & KARKALAS, 1986) and a higher capability of radial pore creation on their surface. This regularity is determined at least for two starchy crops – maize (WANG *et al.*, 1993; GUTIERREZ *et al.*, 2002; GIBBON *et al.*, 2003) and pea (WANG & HEDLEY, 1993; PAVLOVSKAYA, 2001).

Granular sizes, geometrical shape, crystalline structure, chemical qualities and enzyme digestibility of starch granules are genetically determined and differ due to their botanical origin (ANONYMOUS, 1985, 1991; JANE *et al.*, 1994; JANE, 2006) A summary of starch properties of different botanical origin is shown in Table 2.2.2.1.1 (FAO, 1998). Starch granules are also influenced by environmental factors during the period of granule development (BADENHUIZEN, 1963; FREEMAN *et al.*, 1972; BOYER, 1976; CAMPBELL *et al.*, 1996; PENA *et al.*, 2002). In wheat, B-type starch granules have a tendency to be more susceptible to environmental stresses (BLUMENTHAL *et al.*, 1995). With high temperatures after anthesis (37°C day / 28°C night), the class of large A-type starch granules becomes markedly predominant in the mature grains, while small B-type granules are more predominant with lower temperature regimens (HURKMAN *et al.*, 2003). The accumulated temperatures also increase the proportion of amylose in starch (PANOZZO & EAGLES, 1998). Percent volume of wheat starch granules with the size <5.0µm and <9.9µm negatively correlates with the starch and amylose contents in grain, whereas the percent volume of granules with size <2.8µm is positively correlated with the grain protein content (RAEKER *et al.*, 1998). The starch and amylose content in wheat grain is positively correlated with the percent volume of 9.8–18.8µm starch granules (DAI *et al.*, 2008). It was found that the enzymatic hydrolysis rate of waxy, normal, and high-amylose maize starches was positively correlated to the surface area of starch granules (LI *et al.*, 2004).

Numbers of recent reviews are available which integrate current knowledge of starch composition, structure, architecture, functionality, and interactions (WANG, BOGRACHEVA & HEDLEY, 1998; TESTER, KARKALAS & QI, 2004a, 2004b; VANDEPUTTE & DELCOUR, 2004; RUDI *et al.*, 2006; JANE, 2006). As can be seen, structural organisation of the polysaccharide complex of cereal grain is quite complicated, and in spite of numerous special research done (MANNERS, 1989; BALL *et al.*, 1998; ZUGENMAIER, 2003) can be described as still not completely solved.

2.4. BIO-SYNTHESIS OF STARCH STRUCTURAL CO-POLYMERS

2.4.1. Starch synthesis process and its stages

The starch biosynthesis pathway in cereal endosperm is superposed on general metabolism of hexose and hexose phosphate (SMITH, 1999; SCHULMAN, 1999). Briefly stated, the starch synthesis in cereal endosperm consists of the following stages (TETLOW, MORELL & EMES, 2004):

1. the formation of ADP-glucose (ADP-Glc) by ADP-glucose pyrophosphorylase (AGPase) from glucose-1-phosphate and ATP;
2. elongation of the glucan chain by starch synthases;
3. branching of the glucan chain by starch branching enzymes;
4. debranching enzymes are finishing the polymer synthesis.

According to an established view, source substrate for starch structural co-polymers in all starchy crops is sucrose, which during metabolic processes is consequently converted into UDP-glucose, glucose-6-phoshphate, glucose-1-phosphate, ADP-glucose, and only the latest serves as the immediate precursor of amylose and amylopectin synthesis (PREISS & LEVI, 1982; SHANNON & GARWOOD, 1984; CASEY *et al.*, 1993; MARTIN & SMITH, 1995; NELSON & PAN, 1995; LLOYD *et al.*, 1999). It is known that in cereals glucose-1-phosphate is imported into the amyloplast (rather than ADP-glucose or glucose-6-phosphate) which is the substrate for AGPase (KEELING *et al.*, 1988; BOWSHER *et al.*, 1996; SMIDANSKY *et al.*, 2002). Characteristic peculiarity of the starch synthesis process is that its early stages provide only synthesis of the immediate precursor of amylose and amylopectin, namely ADP-glucose. Because of this, effects of genetic factors which regulate activity of sucroso-synthase, phosphoglucomutase and AGPase mainly influence the intensity of starch production, but do not alter distribution ratio of its structural co-polymers (HYLTON & SMITH, 1992; CASPAR, 1994; MÜLLER-RÖBER & KOSSMANN, 1994; HEDLEY *et al.*, 1996; CRAIG *et al.*, 1998; TYMCHOUK *et al.*, 2001; SMIDANSKY *et al.*, 2002).

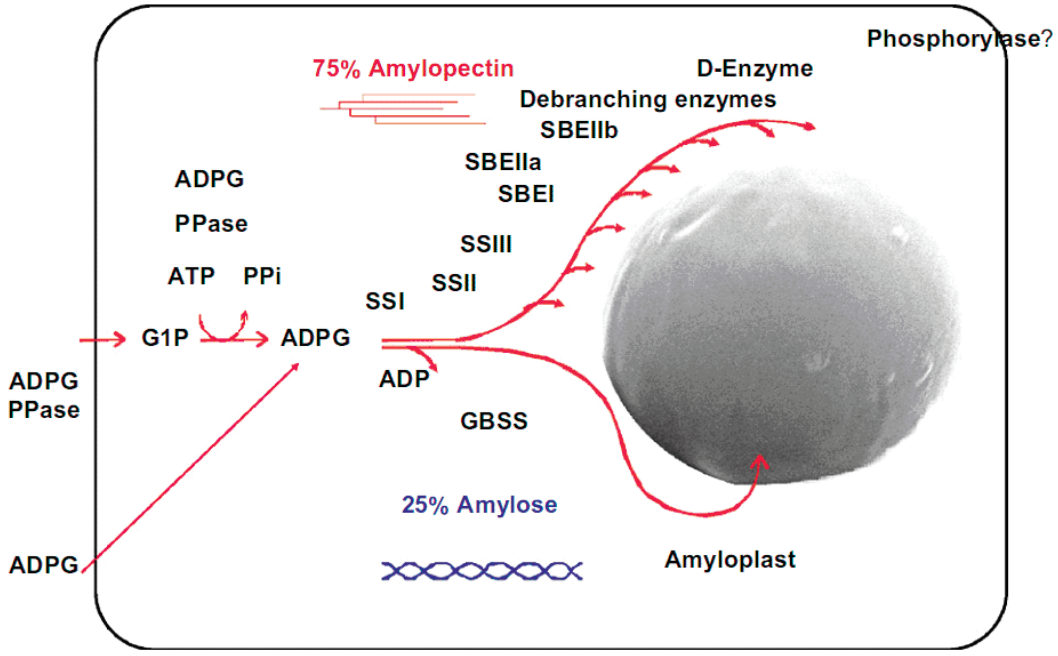
In experiments with pea embryos (DENYER *et al.*, 1996; DENYER *et al.*, 2001) it was shown that malto-olygosaccharides, which are assembled from 2 to 7 glucose

units, are primers for co-polymers of starch synthesis (ZEEMAN *et al.*, 2002). Their addition to isolated starch granules of maize, potato and *Chlamydomonas reinhardtii* had stimulated synthesis of amylose (DENYER *et al.*, 1996; WAL *et al.*, 1998). Elongation of malto-olygosaccharides is accomplished by starch synthases through the process of consecutive addition of isolated glucose molecules from ADP-glucose to maltotriose to maltotetrose, then to maltopentose and so on. This process was named processive elongation (DENYER *et al.*, 1997b).

2.4.2. Catalysis of the starch synthesis and enzymes involved

In the starchy endosperm, a number of enzymatic complexes effects catalysis of various reactions of starch synthesis; some of these enzymes are represented by few isoenzymes (Figure 2.4.2.1; RAHMAN *et al.*, 2007).

Figure 2.4.2.1. Schematic representation of starch biosynthesis in the cereal endosperm (RAHMAN *et al.*, 2007)



In the figure above:
SS I-IV – starch synthases;
GBSS – granule-bound starch synthases;
SBE I-IIb – starch branching enzymes;
D-enzyme – disproportionating enzyme.

These enzymes are represented by sucrose-synthases (SuSy), AGPases, hexokinases, phosphoglucomutases, phosphoglucoisomerases, starch-synthases (SS), starch-branching (SBE) and starch-debranching (DBE) enzymes (SMITH, BETTEY & BEDFORD, 1989; PREISS, 1991; CASEY *et al.*, 1993; SMITH & MARTIN, 1993; HANNAH *et al.*, 1993; BALL *et al.*, 1996; PREISS & SIVAK, 1996, 1998; SMITH, DENYER & MARTIN, 1997; SCHULMAN, 1999; SMITH, 1999). Several detailed reviews had been summarised about recent knowledge on the functional roles of SS, SBE and DBE involved in the synthesis of the starch polymers, as well as starch biosynthesis and its genetic control (RAHMAN *et al.*, 2000; SMITH, 2001; NAKAMURA, 2002; BALL & MORELL, 2003; JAMES, DENYER & MYERS, 2003; TOMLINSON & DENYER, 2003; HILLS, 2004; TETLOW *et al.*, 2004; MORELL & MYERS, 2005; JANE, 2006; TETLOW, 2006).

Plants normally contain multiple isoforms of these enzymes within each class, and all together 14 different isoform classes are found in higher plants (2 APGases, 5 SS's, 3 SBE's and 4 DBE's), 13 of which are homologous in all plants characterised to date (MORELL & MYERS, 2005). Besides these, α-glucan water dikinase (GWD) was shown to be responsible for incorporation of phosphate groups into starch (LORBERTH *et al.*, 1998; RITTE *et al.*, 2002). Fine structure of starch and a quantity of its components may be affected by isoenzymes' different level of functional activity, their concentration, transport, substrate, enzymes interaction, tissular and ontogenetic specificity, spatial and temporal regulation of genes encoding them, host species, as well as biological and macro-environmental conditions (KEELING, BACON & HOLT, 1993; SHI, SEIB & BERNARDIN, 1994; BALL, 1995; MARTIN & SMITH, 1995; WANG, BOGRACHEVA & HEDLEY, 1998; THOMAS & FELL, 1998; FELL, 1999; BÅGA *et al.*, 1999b; BURRELL, 2003; TETLOW, MORELL & EMES, 2004).

AGPase is a main regulator of starch synthesis in plants, which controls the rate-limiting step in starch biosynthesis (SAKULSINGHAROJ *et al.*, 2004; TETLOW, 2006). It catalyses the conversion of glucose-1-phosphate to ADP-glucose, the substrate for starch polymers (amylose and amylopectin) synthesis. It was shown that

AGPase in cereal endosperm is largely extra-plastidial (85–95% cytosolic), but the reverse is true for other cereal tissues and all tissues of non-cereal plants (DENYER *et al.*, 1996; THORBJØRNSEN *et al.*, 1996a; BECKLES, SMITH & REES, 2001; BURTON *et al.*, 2002a; SAKULSINGHAROJ *et al.*, 2004). The cytosolic localisation of AGPase in cereal endosperm has functional significance for partitioning large amounts of carbon into starch when sucrose is abundant (BECKLES *et al.*, 2001). The activity of AGPase is allosterically regulated by inorganic orthophosphate (Pi; inhibitor) and 3-phosphoglyceric acid (3-PGA; activator) (PREISS & SIVAK, 1996; WANG, BOGRACHEVA & HEDLEY, 1998; SIKKA *et al.*, 2001). The barley cytosolic isoform of AGPase is found to be insensitive to allosteric regulation, possessing relatively high activity (RUDI, DOAN & OLSEN, 1997; DOAN, RUDI & OLSEN, 1999). Wheat transgenic plants with mutated maize AGPase genes (which are characterised by increased stability of AGPase subunit interactions and reduced inhibition by Pi) in some cases were shown to have increased seed weight and total biomass (SMIDANSKY *et al.*, 2002). The AGPase is assumed the most heat-liable enzyme in starch biosynthesis in maize (SINGLETARY, BANISADR & KEELING, 1994; DUKE & DOEHLERT, 1996).

It was proposed that plastidic phosphorylase is also involved in starch synthesis (rather than in starch breakdown, as was thought earlier) (DUWENIG, STEUP & KOSSMANN, 1997). Another enzyme, nucleoside diphosphoglucose pyrophosphatase, may be involved in ADP-glucose breaking down, and competing for ADP-glucose as substrate with SS, and thus limiting an overall rate of starch synthesis (BAROJA-FERNÁNDEZ *et al.*, 2000; RODRÍGUEZ-LÓPEZ *et al.*, 2000; KLECZKOWSKI, 2001).

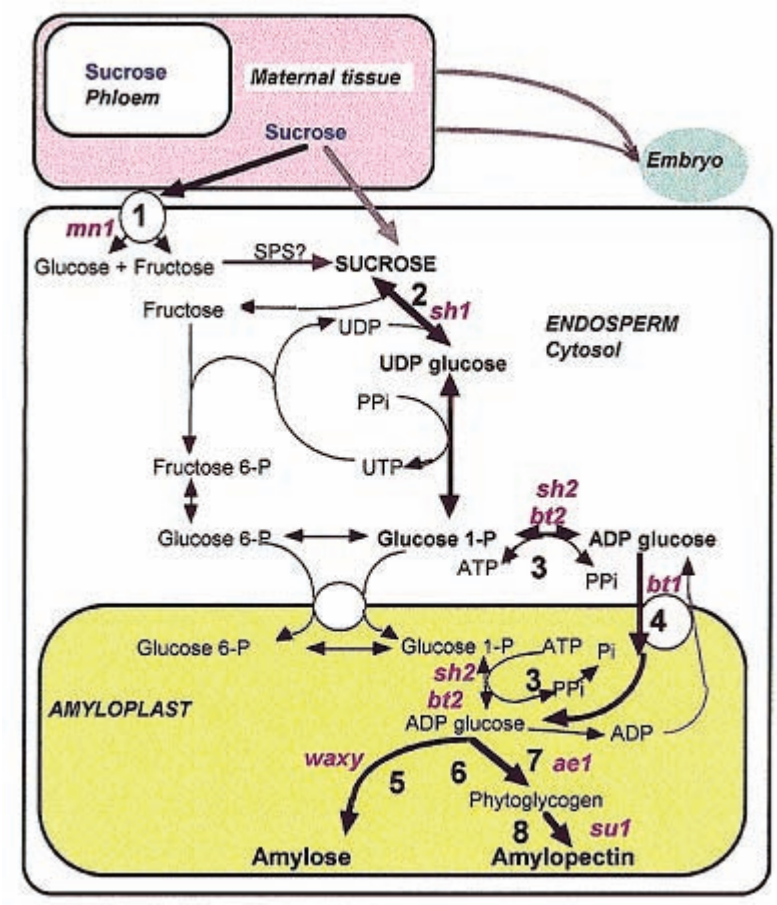
A class of specific 14-3-3 proteins exists that regulates some enzymes activity via formation of complexes with them (CHUNG, SEHNKE & FERL, 1999). The reduction of granule-associated 14-3-3 protein accumulation results in increased starch accumulation. Perhaps it could be explained by the proteins ability of binding with target SSIII that result in inactivation of the enzyme (SEHNKE *et al.*, 2001). Control of the starch synthesis pathway through protein modifications, for instance

regulation of activity of calcium-dependent protein kinase (SPK, which phosphorylates sucrose synthase) was also shown (ASANO *et al.*, 2002).

2.4.3. Amylose and amylopectin synthesis

Immediate synthesis of amylose and amylopectin from ADP-glucose is performed through two independent pathways (Figure 2.4.3.1, SENE *et al.*, 2000; compare with Figure 2.4.2.1).

Figure 2.4.3.1. Present knowledge of the metabolic pathway for starch synthesis in the developing maize kernel (SENE *et al.*, 2000)



In the figure above (enzyme numbering, name and their corresponding mutation in maize):
1 – cell wall invertase / miniature1 (mn1); seems to be required in the basal endosperm, only in the first 10-15 days;
2 – SuSy (sucrose-synthase) / shrunken1 (sh1);
3 – AGPase / shrunken2 (sh2) and brittle2 (bt2);
4 – ADP-glucose translocator / brittle1 (bt1);
5 – GBSS (granule bound starch synthases) / waxy;
6 – SSS (soluble starch synthases);
7 – branching enzymes / amylose-extender1 (ae1);
8 – debranching enzymes / sugary1 (su1).

Amylose synthesis is catalysed by only starch-synthases, but amylopectin synthesis besides starch-synthases is also catalysed by starch-branching and starch-debranching enzymes (WHITT *et al.*, 2002; BALL & MORELL, 2003). For the amylopectin synthesis at least nine isoforms of enzymes is required (JOBLING, 2004). Attempts to produce novel amylopectin types by knockdown of some enzymes in the synthesis pathway did not led to successful results (SAFFORD *et al.*, 1998).

The model of independent synthesis of amylose and amylopectin from ADP-glucose is not exclusive. Some authors acknowledge possibility of amylopectin synthesis from amylose through its extension and branching with help of starch-branching enzymes (TAKEDA, GUAN & PREISS, 1993). There is another model of amylopectin formation from plant homologs of glycogen, which is based on significant similarities of chemical structure of these substances (BALL *et al.*, 1996). The best pretenders for the role of such homologs are branched water-soluble polysucrides, the biggest content of which is registered in sugar maize (INOUCHI *et al.*, 1987; JAMES *et al.*, 1995).

Role of starch synthases (SS). Effect of all SS is in catalysis of the reaction of glucose residue addition from ADP-glucose to non-reducing end of glucan chain, as a result of which the chain elongates through the formation of α-1,4 linkages. Amino acid sequences structure of different isoforms of SS is very similar (BALL *et al.*, 1998; KOSSMAN *et al.*, 1999; LI *et al.*, 1999). All isoforms of the enzyme have three homologous domains and differ from each other by the structure of an N-terminal part, which is located in front of domain I (CORPET *et al.*, 1999). Characteristically, this domain was not found in any other enzyme. Because of temperature sensitivity of SS, as one of the reasons, endosperm filling slows down at temperatures above 20°C, as was shown for wheat (MACLEOD & DUFFUS, 1988; KEELING, BACON & HOLT, 1993; SHI, SEIB & BERNARDIN, 1994; JENNER, DENYER & GUERIN, 1995; BURRELL, 2003).

Conclusions of different authors regarding quantity of SS isoforms that function in cereal endosperm and legume cotyledons do not completely match. Some authors (BOYER & PREISS, 1981; DENYER *et al.*, 1993; MU *et al.*, 1994) hold opinions that soluble fractions of maturing seeds are representing two isoforms of SS plus an extra one, which is localised inside of the starch granules. More recently yet another isoform of soluble SS was identified (DENYER *et al.*, 2001). According to most recent reports, there are at least five isoforms of SS found in cereal endosperm: SSI, SSIIa, SSIIb, SSIII (or SS I-IV, according to some authors) and granule-bound starch synthase (CAO, JAMES & MYERS, 2000; RAHMAN *et al.*, 2007). Each isoform of the SS plays a unique role in amylose and amylopectin synthesis (PREISS & LEVI, 1982; KOSSMAN & LLOYD, 2000). Barley mutants that lacked SSIIa were produced; they had a reduced level of amylopectin and as a result an increased amylose content (MORELL *et al.*, 2003). In wheat, such a high-amylose mutation, deficient in SSIIa (SGP-1) is also known (YAMAMORI *et al.*, 2000). Soluble isoforms of SS mainly secure synthesis of linear polyglycoside sections of amylopectin, when granule-bound starch synthase catalyses synthesis of amylose (SMITH, DENYER & MARTIN, 1995; DENYER *et al.*, 2001; NAKAMURA, 2002).

Granule-bound starch-synthase (GBSS) is represented by two isoforms (DENYER *et al.*, 1997a) and differs from other SS firstly by its localisation. It was suggested that it is located inside of starch granules and is not connected to their surface (VISSER *et al.*, 1991; KUIPERS *et al.*, 1994; RAHMAN *et al.*, 1995; DENYER *et al.*, 1997b; TATGE *et al.*, 1999). Waxy mutants in sorghum were found which produce the GBSS but in a non-functional form (PEDERSEN *et al.*, 2005). Activity of the GBSS increases when granule splitting occurs (HYLTON *et al.*, 1996). Some evidence suggest that GBSS catalyses embedding of glucose not only into amylose, but also into amylopectin and that glucose specifically embed into its longest chain (BABA, YOSHII, & KAINUMA, 1987; WAL *et al.*, 1998). It creates super-long branch chains of amylopectin (YOO & JANE, 2002a; INOUCHI *et al.*, 2005). GBSS-I is absolutely critical for amylose biosynthesis – loss of GBSS-I leads to synthesis of amylose-free (waxy)

starch (FLIPSE *et al.*, 1996; RAHMAN *et al.*, 2000; YANAGISAWA, KIRIBUCHI-OTOBE & YOSHIDA, 2001; GEERA *et al.*, 2005a). Amylopectin in such waxy starch has a greater proportion of longer chains (SASAKI *et al.*, 2002; BERTOLINI *et al.*, 2003).

Amylose synthesis can be regulated by such factors as: amount and existence of different forms of GBSS-I with differential activity; different ontogenetic expressiveness of genetic factors, which control enzyme activity; availability of substrate (ADP-glucose and malto-oligosaccharides) for starch copolymers synthesis; different properties of amylopectin matrix of the starch granule (i.e. availability of physical space within the starch matrix), etc. (RAHMAN *et al.*, 1995; FLIPSE *et al.*, 1996; SMITH, DENYER & MARTIN, 1997; ZHAO *et al.*, 1998; ARAKI *et al.*, 1999; CLARKE *et al.*, 1999; YAMAMORI & QUYNH, 2000; DENYER *et al.*, 2001; EDWARDS *et al.*, 2002; JAMES, DENYER & MYERS, 2003). It was shown that granule-bound proteins with glucosyl-transferase activity are tissue-specific. For instance, such protein in wheat endosperm is a different genetic product than in any other part of the plant (FUJITA & TAIRA, 1998; NAKAMURA *et al.*, 1998).

2.4.4. Isoenzymes involved in synthesis of amylopectin

The role of individual isoenzymes in synthesis of areas with specific branching structure in amylopectin and the determination of its structure and properties was a subject of special studies (MANNERS, 1989; GAO *et al.*, 1998; MYERS *et al.*, 2000). Creation of branching nodes (1,6 links) in amylopectin is catalysed by a specific class of **starch-branching enzymes (SBE)**. Currently there are three known isoforms of this enzyme (i.e. SBEI, SBEIIa and SBEIIb) in wheat (MORELL *et al.*, 1997; NAGAMINE *et al.*, 1997; REGINA *et al.*, 2004; RAHMAN *et al.*, 2001, 2007), maize (BOYER & PREISS, 1978a; FISHER *et al.*, 1993; FISHER *et al.*, 1995; GAO *et al.*, 1997) and rice (MIZUNO *et al.*, 1993, 2001), four in barley (SUN *et al.*, 1997) and at least two isoforms in pea (SMITH, 1988; DENYER *et al.*, 1993; BURTON *et al.*, 1995), haricot bean (NOZAKI *et al.*, 2001; HAMADA *et al.*, 2001) and potato (JOBLING *et al.*, 1999). It was suggested that in the wheat genome many different SBEI genes (up to ten) and

SBEII transcripts (at least three) are present (RAHMAN *et al.*, 1997; MORELL *et al.*, 1997; BÅGA *et al.*, 1999a).

Different isoforms of SBE distinctively differ by their molecular weights and kinetic properties, e.g. in their substrate specificity and length of transferred glucan chain (SMITH, 1988; MIZUNO *et al.*, 1992; GUAN & PREISS, 1993), but all of them are united by two commonalities. Firstly, all SBE isoforms together with soluble SS and perhaps with its granule-bound isoform secure synthesis of amylopectin (DANG & BOYER, 1988; GUAN & KEELING, 1998). Secondly, clearly pronounced differential expression of SBE isoforms at different stages of seed development (BURTON *et al.*, 1995; GAO *et al.*, 1996). Mutations of SBEII that lead to production of starches with a higher amylose level were identified in rice, maize, and pea (BOYER & PREISS, 1981; BHATTACHARYYA *et al.*, 1990; MIZUNO *et al.*, 1993). The SBEI mutant in rice was shown to have a change in amylopectin structure (SATOH *et al.*, 2003). In maize, however, SBEI knockout mutants do not express altered phenotype features (BLAUTH *et al.*, 2002). High-amylose wheat was produced with amylose content of more than 60% by simultaneous inhibition of both SBEI and SBEII enzymes (JOBLING *et al.*, 2003). Similar results were reported for potato, where simultaneous knockdown of SBEI and SBEII allows the resulting amylose level to be higher than 60% (SCHWALL *et al.*, 2000).

In maturing seeds are registered not only processes of starch synthesis, but also its cleavage, which is catalysed by at least three individual enzyme classes. Experimentally proven existence of **starch-debranching enzymes** (**DBE)**, e.g. isoamylases and pullulanases (DOEHLERT & KNUTSON, 1991; JESPERSEN *et al.*, 1993). Most probably, they are responsible for partial breakdown of branching nodes of amylopectin (BALL *et al.*, 1996; ZEEMAN *et al.*, 1998), which may result in synthesis of amylose (WAL *et al.*, 1998). It was suggested that DBE cause ‘preamylopectin trimming,’ which creates an outer layer of short-chained polymers, upon which soluble SS can operate to produce longer chains (BALL, 1995; MOUILLE *et al.*, 1996).

At least three types of isoamylases were found in cereals (KUBO *et al.*, 2005). Loss of functionality in isoamylase-1 (with parallel loss of pullulanase) leads to shrunken grains in rice, barley and maize (*su-1* mutation) (JAMES *et al.*, 1995; NAKAMURA *et al.*, 1996; BURTON *et al.*, 2002b; KUBO *et al.*, 2005). The maize *su-1* isoamylase was found to have an effect on SBEIIa activity (DINGES *et al.*, 2001). It is shown that DBE of isoamylase-type affect starch granules number and their morphology (BURTON *et al.*, 2002b; DINGES *et al.*, 2003). In barley, DBE activity is related to increased proportion of B-type granules (BURTON *et al.*, 2002b). When an activity inhibitor of a pullulanase-type DBE (limit-dextrinase) was down-regulated in barley, it resulted in the reduced content of starch, amylose and small B-type granules. It also led to an altered distribution of glucan chain-length in amylopectin (STAHL *et al.*, 2004; TETLOW, 2006).

Some authors distinguish separate groups of **disproportionating enzymes (DE)** (TAKAHA *et al.*, 1993) and **glucan-water dikinases (GWD)** which are in some way involved in carbohydrate polymers synthesis, although their role is less clear (MORELL & MYERS, 2005). Finally, starch can be also degraded while grain is still developing, via hydrolytic processes that are catalysed by isoforms of amylase (GALE *et al.*, 1983; MORRELL *et al.*, 1995; SUN *et al.*, 1999).

General description of mechanisms of starch structural copolymers synthesis affirms their complex character. Because starch biosynthetic enzymes act as a complex, unexpected genes that influence the formation or function of the complex could also indirectly affect the quality of produced starch (TETLOW *et al.*, 2004). Hereditary fixed re-distribution of starch fractional composition seems to be possible with the precondition of identification of its regulatory genetic factors, genetic diversity in loci of interest, and subsequent targeted breeding and selection.

2.5. GENETIC REGULATION OF POLYSACCHARIDE CONTENT AND ITS FRACTIONAL COMPOSITION IN CEREALS

There are no doubts about the hereditary nature of polysaccharides content and fractional composition in cereal grain. Numerous research studies reported about the similarity of genetic regulatory mechanisms of starch structural copolymers synthesis in crops, which belong to different botanical taxa, including cereals (DRY *et al.*, 1992; NAKAMURA *et al.*, 1993b; HEDLEY *et al.*, 1996; HYLTON *et al.*, 1996). A set of monogenic factors were identified, which control separate reactions of amylose and amylopectin synthesis and the effect of which can be used in breeding. So far, over 20 genes are known which are involved in starch production (NELSON & PAN, 1995; MYERS *et al.*, 2000; WHITT *et al.*, 2002). They are similar in at least four general features (MIKU, 1981; SHANNON & GARWOOD, 1984; PALIJ, 1989), namely:

1. change of synthesis intensity of starch structural copolymers in comparison with normal (non-mutant) genotypes is regulated by recessive (mutant) alleles of these genes, which are in most cases of natural origin;
2. biochemical effects of recessive alleles of all starch-modifying mutations to bigger or lesser extend, but always cause decrease of total starch content in grain;
3. every mutant gene controls activity of only one individual reaction of starch synthesis in its metabolic pathway;
4. mechanism of regulation of these reactions by starch-modifying mutant genes consists in production of functionally passive or low-active isoforms of respective enzymes (down-regulation).

One group of starch-modifying genes catalyses reactions of free sugars interconversion. In maize, for example, gene *sh-1* (*shrunken-1*) belongs to this group, which regulates the activity of glucose-synthase and probably provides more glucose for AGPase (LIANG, ZHANG & CAO, 2001; WHITT *et al.*, 2002); gene *bt-1* (*brittle-1*), which regulates adenylate-traslocating enzyme activity (SHANNON *et al.*, 1996; CAO &

SHANNON, 1997); cytoplasmic AGPase structural genes *sh-2* and *bt-2*, which encode large and small (respectively) AGPase subunits regulated by allosteric effects (BAE *et al.*, 1990; BHAVE *et al.*, 1990; GREENE & HANNAH, 1998b). Mutations of these genes trigger strong reduction in starch content (NELSON, 1982; JOHNSON *et al.*, 2003; SAKULSINGHAROJ *et al.*, 2004).

Another group of starch-modifying mutant genes regulate reactions of synthesis and interconversion of starch structural components, i.e. amylose and amylopectin (JAMES, ROBERTSON & MYERS, 1995). It is the gene *wx*, which regulates the activity of GBSS (SHURE *et al.*, 1983; KLOSGREN *et al.*, 1986), the *ae*, which regulates the activity of SBE (CASEY *et al.*, 1993; FISHER *et al.*, 1993; STINARD *et al.*, 1993), and the *su-1*, which regulates the activity of DBE (JAMES *et al.*, 1995). To this group also belongs gene *du-1*, specificity is that it alters the activity of soluble SS because of SBE activity modification (FERGASON, 1994; GAO *et al.*, 1998).

Starches, which are produced as a result of the effect of different starch-modifying genes, are significantly different from the conventional starch of non-mutant plants and from each other, because of their fractional composition (WANG *et al.*, 1993; KATZ *et al.*, 1993). As was mentioned, the mutant gene *wx* (waxy) has a major effect on high-amylopectin starch synthesis, and on high-amylose starch synthesis – mutant gene *ae* (amylose-extender), which are useful in breeding (FERGASON, 1994). These two mutated genes *wx* and *ae* were utilised in the creation of high-amylopectin and high-amylose genotypes in cereals. Genes with similar effect in relation to high-amylopectin starches synthesis were identified in barley (ROHDE *et al.*, 1988), rice (HIRANO & SANO, 1991), wheat (MURAI *et al.*, 1999), durum wheat (VIGNAUX *et al.*, 2004), pearl millet (*Pennisetum glaucum*; REDDY *et al.*, 1996; RAMESH *et al.*, 1998), sorghum (PEDERSEN *et al.*, 2004), pea (DENYER *et al.*, 1995) and potato (HOVENKAMP-HERMELINK *et al.*, 1987). High-amylose mutations (*ae*) were found in barley (SHONDELMAIER *et al.*, 1992; MORELL *et al.*, 2003), rice (ASAOKA *et al.*, 1986), wheat and aegylops (WATANABE *et al.*, 1998; ARAKI *et al.*, 1999), pea (WANG & HEDLEY, 1993) and potato (SCHWALL *et al.*, 2000). This can serve as

evidence not only of parallelism of genetic variability of crops in starch fractional composition, but also that the practical improvement of these traits can be approached and performed through use of principally similar technologies (NAKAMURA *et al.*, 1995; HEDLEY *et al.*, 1996; REDDY *et al.*, 1996; WATANABE *et al.*, 1998).

In the study with rye MOHAMMADKHANI, STODDARD & MARSHALL (1999a) did noticed that a higher amylose content had a dominant (3:1 ratio) or additive (with 1:1:1:1 or 1:1 ratios) inheritance in different F_2 families, however in one tetraploid family high amylose had recessive inheritance (with 1:3 ratio). They suggested that minor genes and modifiers are also involved in amylose content determination in rye, einkorn (*Triticum monococcum*) and emmer (*Triticum dicoccum*) under their study; cytoplasmic effect was also noticed in reciprocal crosses. The cytoplasmic effect of 0.5–1.5% magnitude on amylose content was reported in rice (POONI, KUMAR & KHUSH, 1993a). Previous studies with rice showed that single dominant or incompletely dominant gene controls its high amylose content (MCKENZIE & RUTGER, 1983; KUMAR & KHUSH, 1986a). The additive nature of genes that control the amylose content in rice was also reported (KUMAR & KHUSH, 1986b, 1988; POONI, KUMAR & KHUSH, 1993a). In research by ARAKI, MIURA & SAWADA (1999) the short arm of wheat chromosome 4A was found to have some quantitative trait loci (QTL) which affects the amylose content. Such QTL's were also reported in rice, which suggests that the trait can be treated as quantitative (CHEN *et al.*, 2008).

Mutants that have the well-known *wx* allele were for a long time employed in production of waxy genotypes, particularly in maize (NELSON & RINES, 1962). Mutant gene *wx* causes the production of starches that are composed almost completely from amylopectin, consisting of 0.00–0.05% amylose (SATHISH *et al.*, 1995; RUDI *et al.*, 2006). Some researchers (YASUI *et al.*, 1996) reckon that starches of such a type can contain up to 1.5–2.0% amylose, but its presence, rather, is linked to the work of DBE. Other researchers showed an even higher amylose content for completely waxy genotypes, with the amylose range (depending on the method of determination) of

0.0–6.0% for wheat, 0.6–2.9% for maize, 4.0–8.4% for barley, and 0.0–2.3% for rice (NAKAMURA *et al.*, 1995; HAYAKAWA *et al.*, 1997; DEMEKE *et al.*, 1999).

The effect of *wx* gene has a fixed character, it is independent from parental genotype combination and weather conditions during plant development, and is inherited as qualitative trait (GRAYBOSCH *et al.*, 2003; VIGNAUX *et al.*, 2004). According to MIURA *et al.* (2002), the *wx* mutation does not cause substantial pleiotropic effect on yield and main grain quality traits in maize and wheat. However, YASUI, SASAKI & MATSUKI (1999) reported reduced (*ca.* 20% lower) flour yield for waxy lines of hexaploid wheat, which was attributed to either reduced content of starch, or higher content of β-glucan, or a higher content of fat. ABDEL-AAL *et al.* (2002) also reported a lower starch content and yield for waxy wheat lines. ROSS, QUAIL & CROSBIE (1996) obtained lower protein content in *Wx-B1b* (mutant) lines compared to wild-type lines. GRAYBOSCH *et al.* (2003) reported that protein-quality traits of waxy bread wheats were not widely deviating from non-waxy check cultivars, and that starch-related characteristics were relatively stable across different environments. For durum wheat specifically, it was shown that grain yield, whole grain protein content, kernel size, or kernel hardness were not affected by waxy mutation, but had increased α-amylase activity and ash content in the whole grain (SHARMA *et al.*, 2002; GRANT *et al.*, 2004; VIGNAUX *et al.*, 2004).

Isoforms of GBSS-I (the *wx* proteins) are found in different cereals, including maize (KLOSGREN *et al.*, 1986), barley (ROHDE *et al.*, 1988) and rice (WANG *et al.*, 1990). There are no reports on mutations in any cereal leading to the over-production of GBSS-I that could lead to increased amylose synthesis. *Wx* proteins in hexaploid wheat are encoded by three homologous *wx* genes (*Wx-A1, Wx-B1,* and *Wx-D1*) located on three chromosomes 7AS, 4AL, and 7DS, respectively (NAKAMURA *et al.*, 1993a; FUJITA *et al.*, 1996; YAN *et al.*, 2000). Genome-specific primers for *wx* genes of wheat are available (BLAKE *et al.*, 2004). Effects of these three *wx* isoforms in wheat on amylose content are different from each other, and in addition, the hexaploid nature of a wheat genome has a buffering effect on these *wx* gene expressions

(YAMAMORI *et al.*, 1994; MIURA & TANII, 1994; SVIHUS, UHLEN & HARSTAD, 2005; HUNG, MAEDA & MORITA, 2006). Ranges of amylose content in wild-type and single null *wx* wheat overlap, and the combination of two null *wx* alleles reduces amylose content only by about 5%, compared to that in wild-type (GRAYBOSCH, 1998). The influence of *wx* genes on amylose deficiency is expressed differentially, with the biggest influence being when *Wx-B1* protein is absent, followed by a lack of *Wx-D1*, and the smallest influence expressed by the *Wx-A1* deficiency (MIURA & SUGAWARA, 1996; MIURA *et al.*, 1999, 2002; ARAKI, MIURA & SAWADA, 2000; YAMAMORI & QUYNH, 2000; MANGALIKA *et al.*, 2003; OKUNO, 2005). The effect of different mutant *wx* gene combinations on starch properties is realised through changed amylose content and its localisation within starch granules (KOZLOV *et al.*, 2006). When the *wx* gene dosage was increased in tetrasomic lines (which carried either of the three *wx* genes by 6 dose), it did not increase amylose content above 25% despite increased GBSS-I activity (MIURA & SUGAWARA, 1996; WICKRAMASINGHE & MIURA, 2003). The degree of branching or chain length of amylopectin is not affected by the reduced GBSS-I activity introduced by null *wx* alleles (MIURA *et al.*, 2002). When other enzymes are simultaneously affected by a mutation, it can also alter the amount and branching pattern of amylopectin (RAHMAN *et al.*, 2007).

The opposite effect of starch copolymers re-distribution is caused by mutant genes *ae*, *su*, and *du* (YAMAMORI *et al.*, 2000; RAHMAN *et al.*, 2007). Mutants in locus *ae*, which encodes SBEIIb enzyme, were successfully utilised for the creation of high-amylose genotypes in barley, maize and rice (BOYER & PREISS, 1978b). Locus *ae* has multiple allelism, i.e. different combinations of alleles of the locus *ae* control different isoforms of SBE, as was shown in maize (FISHER *et al.*, 1996; KIM *et al.*, 1998). Starches that are synthesised by means of the control of different alleles of locus *ae* are non-identical in their relative amylose content, their degree of conversion of amylopectin into maltose, and their disposition of iodine absorption complexes (GARWOOD *et al.*, 1976). In maize loss of functionality in SBEIIb leads to production of up to 80% amylose in starch (SHANNON & GARWOOD, 1984). Alike mutation in rice

leads to an increase in amylose content of up to 35% (NISHI *et al.*, 2001). Protein SGP-2 in wheat is homologous to maize SBEIIb enzyme (FISHER, BOYER & HANNAH, 1993). In wheat, SBEIIa is known to be the predominant isoform, and a natural high-amylose mutation of it is not known so far (RAHMAN *et al.*, 2007). However, the genetic modification approach was successfully used for the simultaneous knockout of SBEIIa and SBEIIb to produce wheat with about 80% amylose content (REGINA *et al.*, 2006). Mutations in SBEI do not seem to alter phenotypes of wheat (even triple null) and maize (BLAUTH *et al.*, 2002; REGINA *et al.*, 2004). However, in maize a combination of inactive SBEI and SBEIIb leads to production of higher branched starch without any change in amylose to amylopectin proportion (YAO *et al.*, 2004). Rice mutation that eliminates SBEIIb activity is shown to interact with the SSI enzyme, activity of which is also reduced by 50% in such mutants (NISHI *et al.*, 2001).

SSI, SSII and SSIII enzymes are responsible for the length of the growing amylopectin chains (JAMES *et al.*, 2003). SSI mutation in rice does not affect the amylose/amylopectin ratio, but alters the starch structure by increasing the proportion of short chains (DP 6–7) and long chains (DP 16–19), and reducing the proportion of DP8–DP15 chains in amylopectin (FUJITA *et al.*, 2006). SSI mutation in barley (at *shx* locus) leads to lower SSI activity and an associated reduction of the size of A-type starch granules, thus making them of a unimodal size with B-type granules (SCHULMAN & AHOKAS, 1990; TYYNELA & SCHULMAN, 1993; TYYNELA *et al.*, 1995). Wheat protein SGP-3 is known to be a homolog of maize SSI enzyme (KNIGHT *et al.*, 1998). Lack of the wheat protein involved in starch synthesis, SGP-1, which is bound exclusively to starch granules (DENYER *et al.*, 1995; RAHMAN *et al.*, 1995; LI *et al.*, 1999), was shown to be responsible for enhanced apparent amylose content and altered amylopectin (YAMAMORI *et al.*, 2000).

When wheat completely lacks the SSIIa enzyme (triple mutant), the average amylopectin chain length is reduced, and the amylose content increases to about 35% (YAMAMORI *et al.*, 2000; HUNG, MAEDA & MORITA, 2006). In barley, mutation that is responsible for the absence of the SSIIa enzyme leads to an increase in amylose

content of up to 65% (MORELL *et al.*, 2003). However, this mutation (in both wheat and barley) also indirectly affects other enzymes, so it is difficult to distinguish the exact cause of such a shift in amylose synthesis (MORELL *et al.*, 2003; KOSAR-HASHEMI *et al.*, 2007). In maize, mutation in SSIIa is associated with an increase in the amylose content from 26 to 40% and is called *su-2* (*sugary-2*) (ZHANG *et al.*, 2004). In rice, the difference between sub-species *indica* and *japonica* lies in the lack of the SSIIa enzyme in *japonica*-type rice (UMEMOTO *et al.*, 2002). Nevertheless, this difference does not lead to a higher amylose content in *japonica* rice, as would be expected, and this is because of the higher activity of the GBSS enzyme in *indica* rice, which ultimately leads to a higher amylose content in *indica* than in *japonica* rice (HIRANO *et al.*, 1998; RAHMAN *et al.*, 2007). Loss of SSIII activity in maize is expressed as *du-1* (*dull-1*) phenotype, which results in a moderate increase of amylose content (GAO *et al.*, 2001; TZIOTIS *et al.*, 2004). SSIV is known to be produced in grain during its development; however, so far no mutations in SSIV were reported for major crops (HIROSE & TERAO, 2004; DIAN, JIANG & WU, 2005; RAHMAN *et al.*, 2007).

Because specific functions are assignable to all individual enzyme isoforms (MORELL & MYERS, 2005), it is possible to obtain a wider genetic variation in starch composition by looking for mutations in genes by which they are encoded. Plant material with such a wide range of starch properties could be exploited by means of breeding plants to satisfy requirements of different industries (RUDI *et al.*, 2006), including fermentations for bio-ethanol production.

2.6. TECHNOLOGICAL PROPERTIES AND INDUSTRIAL APPLICATIONS OF HIGH-AMYLOSE AND HIGH-AMYLOPECTIN STARCHES

2.6.1. Factors which determine technological properties of starch

Approximately 70% of all the starch produced is used in the food industry as a thickening, stabilising, and gelling agent (SLATTERY, KAVAKLI & OKITA, 2000;

TESTER & KARKALAS, 2002). The remaining 30% of industrial starch applications include: an additive in cement to improve its setting time; the improvement of the viscosity of drilling solutions used in oil wells; in paper-making as a filler that bonds the cellulose fibres together and improves the strength of the paper. It is also used as an adhesive in paper bags, and as a stilt base in carbonless copy papers (WHITE, 1998; BURRELL, 2003).

The technological properties of starch are characterised by numerous combinations of independent traits, but leading among them is the ability of starch granules to swell, the stability of the starch molecular structure during the disperse phase, the gelling ability of the starch, and its suitability for digestion by amylolytic enzymes (RIKHTER *et al.*, 1975; KNUTSON *et al.*, 1982; ZIEGLER *et al.*, 1993; FISHER & THOMPSON, 1997). Starches gelatinise when there is an excess of water and when they are heated to between 50–80°C, when nearly all amylose in the starch granule leaches out; this process is irreversible (HAN & HAMAKER, 2001). Smaller starch granules require a higher temperature for gelatinisation (VASANTHAN & BHATTY, 1996; CHIOTELLI & LE MESTE, 2002). Experiments regarding the ability of high-amylose and high-amylopectin starches to swell and gelatinise at different temperatures showed contradictory results (YASUI *et al.*, 1996; HAYAKAWA *et al.*, 1997; FUJITA *et al.*, 1998; VASANTHAN & BHATTY, 1996; CHIOTELLI & LE MESTE, 2002; HUNG, MAEDA & MORITA, 2006). Starch gelatinisation leads to a loss of the crystalline structure and consequently leads to increased susceptibility for amylolytic degradation (KISHIDA *et al.*, 2001; SVIHUS, UHLEN & HARSTAD, 2005). It was found that a correlation of 0.96 exists between the extent of gelatinisation and the digestion rate of pure starch (HOLM *et al.*, 1988).

Due to the re-distribution of fractional composition and their morphology alteration, starches that are synthesised by different mutant genes essentially differ from each other in terms of their technological properties (BROCKETT *et al.*, 1988; KATZ *et al.*, 1993).

2.6.2. Properties and application of high-amylopectin starches

Research done on high-amylopectin and completely waxy starches (YUAN *et al.*, 1993; LIU & THOMPSON, 1998; SEMEIJN & BUWALDA, 2006) was able to establish that gene *wx* provides for the production of starches that are distinguished by their low temperature of gelatinisation, fast hydration, transparency, high water binding ability, increased viscosity, and salt stability of gels. Additionally, high-amylopectin starch, with its increased availability for amylolytic enzymes, can easily be digested and thus rapidly assimilated by living organisms (SANDSTEDT, HITES & SCHROEDER, 1968).

These qualities provide for diverse opportunities in the use of high-amylopectin starch as a thickener, emulsifier, glue material and as a valuable component of dietetic and children's nutritional products. Perspective areas of application of such starches is in the production of concentrated soups, puddings, custard, jelly, fruit and milk desserts, various sauces and pastes (WHITE, 1994; SHARMA *et al.*, 2002; ISHADA *et al.*, 2003). Waxy starches improve the processing quality, palatability, and shelf life stability of baked and sheeted wheat products (LEE, SWANSON & BAIK, 2001; HAYAKAWA *et al.*, 2004; SAHLSTRÖM, BÆVRE & GRAYBOSCH, 2006). Its application is important in processed meat products as a binder that maintains stability and texture (BURRELL, 2003). Because of the spreading of metabolic diseases, caused by a breach in the metabolism of amino acids, high-amylopectin starches are used as structural components of protein-free foodstuffs (ZHUSHMAN *et al.*, 2001). In the pharmaceutical industry, high-amylopectin starches have considerable possibilities in obducing, tabletted, and emulsifying applications during pharmaceutical production (MURAV'EVA, 1991; STEFFENS, 2006), and in the mining industry – as a component of drilling solutions (SHITS, 2001).

2.6.3. Properties and application of high-amylose starches

High-amylose starches have completely different technological properties, compared to those of high-amylopectin starches (BOLTZ & THOMPSON, 1999; KLUCINEC & THOMPSON, 1999; RICHARDSON, JEFFCOAT & SHI, 2000; SUH *et al.*,

2001). They form hard, dense gels with a high strength of extension and an elastic structure, which is useful in the production of sweets. Amylose acts as an inhibitor of swelling, especially in the presence of lipids (TESTER & MORRISON, 1990; LII, TSAI & TSENG, 1996), and contributes to an increase in pasting temperatures (JANE & CHEN, 1992). Such properties are much desired for the production of some specific types of foodstuff, such as starchy noodles and special 'resilient' types of baked products (BIRD, BROWN & TOPPING, 2000). High-amylose starches are also used for the production of high-quality photo and cine film (SUH *et al.*, 2001) and are considered to be the best raw material for the production of biodegradable thermoplastics (GRIFFIN, 1989; COLONNA *et al.*, 1995; KALUGINA & ZAPOL'SKAYA, 2001; RINDLAV-WESTLING, STADING & GATENHOLM, 2002; WEBER *et al.*, 2002; ALI, 2002). High-amylose starches containing small granules are required for the plastics industry (WANG, BOGRACHEVA & HEDLEY, 1998).

High-amylose starches demonstrate increased resistance to amylolytic hydrolysis (SANDSTEDT *et al.*, 1962; HUNG, YAMAMORI, & MORITA, 2005), thus increasing the level of glucose in the bloodstream to a much lesser degree than common starches (CHAMP & NOAH, 1996). Hence, they are considered as perspective raw materials for foodstuffs with hypoglycaemic activity, resulting in improved nutritional value (THOMPSON, 2000; NIKOL'SKAYA *et al.*, 2001; HUNG *et al.*, 2008).

Therefore, starches of both types – high-amylopectin as well as high-amylose – certainly can be seen to have possibilities in practical applications. Generally, efforts have been concentrated on breeding for increased starch yield (for food, feed, and ethanol industries). Yet, the creation of cereal cultivars with high-amylose and high-amylopectin starches, and their modifications that are characterised by improved properties (DAVIS *et al.*, 2003), can be viewed as independent directions of improvement of starchy crops by means of breeding.

2.7. ANALYTICAL METHODS OF GRAIN COMPONENTS DETERMINATION

Selection of genotypes with increased starch content requires simple, yet repeatable methods of desirable genotypes identification (HUCL & CHIBBAR, 1996). There are direct analytical chemistry methods for the determination of starch and its components, which are used as a reference for derived instrumental methods such as near infra-red spectroscopy. In general, direct measurements are used to provide accurate estimates of quality attributes, and indirect methods (derived/calibrated from the direct measurements) were developed for rapid use at point of production, for trade, or in plant breeding.

2.7.1. Direct methods of starch determination

Starch is known to be notoriously difficult to measure, with different analytical methods giving substantially different results (SMITH *et al.*, 2006). Most of the quantitative methods of starch analysis are based on a two-stage procedure, the first step being the hydrolysis of starch into glucose by either acid or enzymes, followed by quantification of the produced glucose. Analytical methods of glucose measurement include polarimetry, colorimetry, gas-liquid chromatography (GLC), and high performance liquid chromatography (HPLC). A highly sensitive method for determining low starch content materials and for micro-scale analysis was developed; this method uses direct determination of glucose after starch hydrolysis. It is based on high performance anion exchange chromatography and pulsed amperometric detection (HPAEC/PAD) (LEVINE, BAUER & LEVINE, 2005).

The main methods employed for starch analysis in commercial practice are described below (after SMITH *et al.*, 2006).

2.7.1.1. Hydrolysis methods employed during starch analysis

Acidic hydrolysis. Diluted mineral acids (most often 1M sulphuric acid which is fairly selective for starch) can be used for starch solubilisation and hydrolysis. This procedure effectively gelatinises the starch granules and simultaneously hydrolyses

the starch to glucose in a single step. In the Ewers' (polarimetric) method (USCL method 11-02, and ICC Standard No. 123/1, http://www.icc.or.at/methods3.php), the starch is released from milled wheat flour by boiling it in diluted hydrochloric acid (HCl). However, there is always some loss of sugars during acidic hydrolysis (about 10–20%, depending upon time, temperature, and acid concentration). Thus, a correction factor has to be applied when the actual starch concentration is estimated. Another method uses a non-acidic hydrolysis of starch content by means of hot calcium chloride dissolution (ICC Standard No. 122/1, http://www.icc.or.at/methods3.php).

Enzymatic hydrolysis. Modern methods of analysis use enzymes rather than acid for starch breakdown. The advantage is that there should be no loss of glucose through degradation, as in the case of acidic hydrolyses. Three key enzymes are needed for the complete breakdown of starch: α-amylase and pullulanase – for the breakdown of the α-1,6 branching points of amylopectin and amyloglucosidase – for breakdown of any residual glucans and maltose to glucose. Addition of pullulanase can be omitted because α-amylase can also break down α-1,6 branch points of amylopectin, although slowly. Thermostable α-amylases that can work at 95°C are normally employed (RICHARDSON *et al.*, 2002), which is very important because of the high temperature helping gelatinisation and solubilisation of the starch. This is critical in ensuring complete starch hydrolysis. Insufficient gelatinisation and dispersion is the most common source of inaccuracy in enzyme-based starch determinations, which leads to underestimating the true starch content.

2.7.1.2. Quantification methods of the produced glucose

Polarimetric measurement of glucose for starch analysis. In the polarimetric (or Ewers') method, glucose concentration is determined by measurement of the polarisation angle or by the optical rotation of the solution (SENN & PIEPER, 2000). Because of the natural presence of free sugars in whole grain, their content has to be measured separately and subtracted from the total glucose

estimation. An additional portion of free sugars is generated from NSP after their acid hydrolysis, which can contribute to an error in the glucose quantification and needs to be taken into account. For this reason, the Ewers' method is stated to be an unsuitable method for samples that are expected to have high levels of NSP or optically active substances that do not dissolve in 40% ethanol (VAN EYS, OFFNER & BACH, 2004). The polarimetric method is of little value for cereal flours and samples with a relatively low starch content, where an enzymatic method has to be employed (MCCLEARY, SOLAH & GIBSON, 1994).

Colorimetric (enzyme-based) measurement of glucose for starch analysis. Glucose released during enzymatic digestion can be measured using GLC and HPLC, which are lengthy and/or expensive techniques and thus are not practical for routine use. Enzymatic methods exist that employ specific enzymes for the oxidation or phosphorylation of glucose, with the consequent measurement of the reaction products by means of a spectrophotometer (HOLM *et al.*, 1986; MORALES, ESCARPA & GONZALEZ, 1997; NEBESNY, ROSICKA & PIERZGALSKI, 1998; BRUNT, 2000). Normally such methods employ glucose oxidase/peroxydase (GOPOD) reagent, which gives a colour reaction with glucose, to determine glucose concentration during the final stage of testing (AACC method 76-11 and its modifications AOAC 996.11 and AACC 76-12, see MCCLEARY *et al.*, 1997; MCCLEARY, SOLAH & GIBSON, 1994; DEMIATE, KONKEL & PEDROSO, 2001). Enzyme-based assay kits are available on the market (MCCLEARY *et al.*, 1997). These kits are of a high purity and selectivity, robust, yet only reliable in the hands of a skilled regular operator, are laborious, more complex, and relatively expensive per sample tested (SMITH *et al.*, 2006). In these enzymatic methods, error contributing factors mainly flow from inappropriate sampling and tissue preparation protocols, as well as: incomplete removal of interfering soluble sugars before starch hydrolysis; non-specific hydrolysis during gelatinisation of starch granules; incomplete hydrolysis of starch due to insufficient amounts of hydrolysing enzyme; improper use of starch standards (LEVINE, BAUER & LEVINE, 2005).

2.7.2. Alternative (indirect) methods of carbohydrates estimation in grain

As an alternative and perhaps better approach to the determination of grain quality for ethanol production, protein measurement can be used instead of starch determination, because protein and starch contents in grain are in a strongly negative correlation. Protein measurement has a few definite advantages over starch determination methods (SMITH *et al.*, 2006):

1. protein determination methods have a significantly higher degree of precision than starch determination methods;
2. protein and ethanol yield have a clearly inverse relationship;
3. the relatively inexpensiveness of protein analysis;
4. protein content can be more easily predicted and controlled by agronomic practice, specifically nitrogen fertiliser inputs.

There are two methods used for protein determination in grain, both based on the measurement of nitrogen content, namely the Kjeldahl and Dumas methods (AACC, 2000). The actual protein content is then determined by the multiplication of nitrogen content by a constant, according to the botanical origin of the grain (MOSSÉ, 1990; IDF, 2006; SMITH *et al.*, 2006).

2.7.3. Amylose content measurement

Amylose content has historically been determined by the iodine-binding procedure of either amperometric, potentiometric or spectrophotometric detection (CHEN & BERGMAN, 2007). Structural co-polymers of starch are clearly distinguished by their characteristic iodine-starch reaction, in which the iodine solution colours the amylose blue, and the amylopectin a reddish-violet (PEDERSEN *et al.*, 2004). Amylose colouring is the result of the creation of a complex chemical compound, in which iodine atoms are located on the inside of amylose chain spirals. Regarding amylopectin, its colouring by iodine results from the creation of adsorption compounds (DENYER *et al.*, 2001). A rapid method for the screening of plant breeding

material was developed based on iodine staining of pollen and grain, which allows determination of waxy genotypes and phenotypes (PEDERSEN *et al.*, 2004). However, iodine also binds with the amylopectin with DP>60, which causes an overestimation of the amylose content (WANG, BOGRACHEVA & HEDLEY, 1998). Furthermore, the phospholipids and free fatty acids compete with iodine in the formation of complexes with amylose, which tends to cause underestimation of the amylose content. Consequently, amylose content measured using iodine-based methods has been termed 'amylose equivalents' or 'apparent amylose' (TAKEDA, HIZUKURI & JULIANO, 1987). The operator and laboratory-dependence of these iodine-binding methods have been reported in literature (BATEY & CURTIN, 1996). These methods are prone to inter-laboratory variability because of the complexity of the procedure and its reliance on amylose and amylopectin standards for the establishment of reference curves (DELWICHE, MCKENZIE & WEBB, 1996). Each method is briefly described below.

Potentiometric or amperometric titration of bound iodine methods (BANKS, GREENWOOD & MUIR, 1971). The method is also based on the inherent capacity of amylose to accommodate poly-iodide ions, mainly pentaiodide anion I_5^-, within its helical structure, with amylopectin lacking such capacity (HIZUKURI, 1996). The potentiometric method suffers from it being a slow, non-ionic reaction, and the broad inflection point leads to some inaccuracies coming to the fore (MCGRANCE, CORNELL & RIX, 1998).

Colorimetric (spectrophotometric) methods. After the iodine has formed complexes with amylose and amylopectin, the absorbance of the blue-coloured amylose-iodine complex is measured, which allows for the determination of the iodine-binding capacity of starch ('blue-value') (MARTINEZ & PRODOLLIET, 1996). Normally the method gives a good correlation (near 96%) between the blue-value measurements of iodine complexes and the amylose content determined directly by size-exclusion chromatography (KNUTSON & GROVE, 1994). The method requires measurement at only one wavelength (e.g. 600nm) and avoids the use of harsh dispersants for the starch (KNUTSON & GROVE, 1994; MCGRANCE, CORNELL & RIX,

1998). It is the most economical and most rapid, and hence probably the most commonly used method (MOHAMMADKHANI, 2005). It is attractive because of its versatility and simplicity; samples of high and low amylose content may be analysed. The sensitivity of the iodine reaction is quite high – it is applicable to amounts of starch which contain as little as 100mg of amylose (MCGRANCE, CORNELL & RIX, 1998). However, the method is not perfectly accurate due to interference from amylopectin, and molecules with degree of polymerisation and structure that is intermediate between amylose and amylopectin (WANG, BOGRACHEVA & HEDLEY, 1998). It is subject to interference from lipids bound to the amylose, and the true amylose content is determined only after a lengthy de-fatting process, without which it is more correct to describe the result of the measurement as an 'apparent' amylose (MOHAMMADKHANI, 2005). It is important to note that various modifications of the method were developed (LUSTINEC *et al.*, 1983; MIURA *et al.*, 1994; MARTINEZ & PRODOLLIET, 1996; MOHAMMADKHANI *et al.*, 1999b).

2.6.3.1. Amylose content measurement – non-iodine methods

Differential scanning calorimetry (DSC) is based on the formation of a complex between amylose and added excess phospholipids (L-α-lysophosphatidylcholine from egg yolk) and its resulting change in enthalpy during cooling (MESTRES *et al.*, 1996). The DSC method appeared to have an improved repeatability compared to the iodine-binding method (POLASKE *et al.*, 2005), and the DSC method is not influenced by the presence of amylopectin or indigenous lipids (MESTRES *et al.*, 1996). This method is expected to be effective in discriminating high-amylose genotypes in a breeding programme specifically designed to identify maize starches with amylose values at or above 70%.

Size exclusion chromatography (SEC), high-performance liquid chromatography (HPLC) – these methods are used to determine amylose content by quantification of the amount of amylose relative to amylopectin (GRANT, OSTENSON & RAYAS-DUARTE, 2002; CHEN & BERGMAN, 2007). SEC methods were reported to

be superior due to their ability to determine absolute amylose content, the lack of interference from lipids, and their power to separate amylose and amylopectin based on differences in their hydrodynamic volume (CHEN & BERGMAN, 2007).

The **lectin-binding method** of amylose content determination was developed using a lectin, namely concanavalin A, that interacts specifically with α-D-glucosyl units of amylopectin at multiple non-reducing end-groups and as a result forms a precipitate with amylopectin (GIBSON, SOLAH & MCCLEARY, 1997). Remaining carbohydrates (i.e. amylose) in the solution are digested by enzymes to form glucose, which is consequently measured by spectrophotometry. Advantages of this modified concanavalin A procedure for amylose determination include its applicability to flour samples without the need for prior starch purification; it allows the simultaneous estimation of total starch, and does not require a calibration curve (no need in amylose standards). Comparable results in measured amylose content were reported when SEC, iodine-binding, DSC and lectin-binding methods were compared (BATEY & CURTIN, 1996; GERARD *et al.*, 2001).

2.7.4. Instrumental grain composition measurements – near infra-red reflectance/transmittance (NIR/NIT) spectroscopy

Near infra-red reflectance/transmittance spectroscopy (NIRS/NITS) is an established instrumental technique which in the past 30 plus years became widely used in the pharmaceutical, petrochemical, agricultural and food industries. It can rapidly (in seconds, and even continuously in real-time monitoring) provide both physical and chemical information about a given sample. It is able to simultaneously measure a number of parameters, for instance protein, moisture, fibre, oil, digestible energy, starch, and amylose content in grain (BARTON & WINDHAM, 1988; OSBORNE, FAERN & HINDLE, 1993; DELWICHE *et al.*, 1995; SEKULIC *et al.*, 1998; WILLIAMS & NORRIS, 2001; MILLER, 2001; MARK, 2001; WU, SHI, & ZHANG, 2002; STAERK & GRUNEWALD, 2006; BRUNT, 2006). Standard NIR-based methods were approved – see ICC Standard No. 159 “Determination of Protein by Near Infrared Reflectance (NIR)

Spectroscopy"; ICC Recommendation No. 202 "Procedure for near infrared (NIR) reflectance analysis of ground wheat and milled wheat products" (http://www.icc.or.at/methods3.php).

Advantages of NIRS/NITS compared to conventional chemical laboratory methods (HIUKKA, 1998; MIRALBES, 2004; DOWELL *et al.*, 2006b; OSBORNE, 2006; LU, HUANG & ZHANG, 2006) include:

1. Minimal or no sample preparation, simple and rapid measurement – after calibrations are developed and installed, the NIRS/NITS system requires little user skill and attention, and has high speed.
2. Simultaneous determination of multiple traits – a wide range of parameters determined from one assessment of a particular sample, concurrent analyses of multiple constituents from one spectrum.
3. Non-destructive – materials can be shipped and reused after measurements, therefore this method can often be used on a whole grain. This is of particular value in plant breeding because the preservation of seeds, after the initial measurement has taken place, for further analysis or for propagation is important (VELASCO, GOFFMAN & BECKER, 1999; BAYE & BECKER, 2004). However, it is not always possible to use the analysis of whole grain due to the accuracy of results not being as desirable as those from flour samples. Calibrations developed from flour spectra seem to be superior to calibrations derived from whole grain samples (BAO, CAI & CORKE, 2001; SOHN *et al.*, 2004; BAO, SHEN & JIN, 2007; BAO, WANG & SHEN, 2007a).
4. Spectroscopy methods can be used in remote sensing of alive plant canopies (CURRAN *et al.*, 1992).
5. Can provide real-time results, which allow for corrective actions to be applied in on-line quality control systems.
6. Is adaptable to existing automated sorting technologies (DELWICHE *et al.*, 2006).

7. Is more flexible – can be applied for measurements of all kinds of sample types, and only a small amount of material is required for analysis.
8. Sub-sampling is not necessary – not only a part of a population, but a whole population can be analysed because of the high-throughput and flexibility of the method.
9. Economical – low cost per test, no need for reagents and/or manpower.
10. Environment-friendly – it is clean because no wastes emerge.

Near infra-red spectroscopy methods employ illumination of samples with light in near infra-red diapason and measurement of the amount of light absorbed by the sample. It is based on the principle that light is absorbed proportionally to the concentration of chemical bonds in a material (PAULSEN & SINGH, 2004), and thus by recording specific absorption patterns in the near infra-red region the chemical composition of the material being analysed can be reported (WILLIAMS & NORRIS, 2001). Samples are illuminated at different infra-red wavelengths that allow quantification of near infra-red energy absorption by various chemical components at specific wavelengths and at reference wavelengths (MILLER, 2001; LU, HUANG & ZHANG, 2006). For instance, principal absorption bands of water are present at (approximately) 760, 970, 1190, 1450 and 1940nm, of protein at 985, 1140, 1185, and 1435nm, and of starch at 985, 1180, 1440 and 1650nm, thus allowing determination of moisture, protein and starch contents in a product with near infra-red spectroscopy (BEN-GERA & NORRIS, 1968; WILLIAMS, 2001). The near infra-red energy absorption results mainly from the stretching and deformation of N–H linkages between amino acids of protein molecules, O–H bonds of water and carbohydrates, and C–H bonds of fat or oil. After mathematical processing of the raw spectral data for background correction and outliers detection (NAES & ISAAKSSON, 1992; PIERNA & DARDENNE, 2007), the proportion of the absorbed light at specific wavelengths is correlated to the amount of a particular component (e.g. starch, protein, moisture, etc.) in the sample.

In order to measure the substance of interest by NIRS, the relationship between the reflectance of the sample and the amount of the substance of interest has to be established by extensive chemometrics multivariate data processing (such as principal components analysis (PCA), artificial neural networks (ANN), multiple linear regression (MLR), partial least squares (PLS) regression), since many chemical compounds have broad and strongly overlapping spectra in the NIR region (MARTENS & NAES, 1991, 2001). It is done via the development of a multivariate calibration model between the amounts of substance determined by a reference (i.e. wet-chemistry) analytical method and the reading of NIR spectra, by employment of special software for multivariate data analysis (ESBENSEN, 2002). Thus, analysis by NIRS is dependent on a reliable calibration against a suitable standard method. Sometimes NIRS calibrations can be more reliable than other analytical methods, as was demonstrated in the case of digestible energy concentration prediction in compound diets, where NIRS equations were shown to be more accurate than equations based on chemical composition, digestible nutrients or in vitro digestibility (XICCATO *et al.*, 2003).

The robustness and precision of an NIRS method depends on two main factors (SMITH *et al.*, 2006):

1. **The number and nature of the reference samples**. Reliable calibrations require a large number of samples covering the full range of genotypes and environments from which the NIRS laboratory will be receiving samples for analysis in the future. This implies use of a typical set of samples with the widest possible range of the substance of interest (e.g. starch) from the target region/environment for the calibration development (WILLIAMS & NORRIS, 1987). Sample representativeness, sampling procedure and sample preparation method is also important (LU, HUANG & ZHANG, 2006; BAO, SHEN & JIN, 2007). Better results (but not necessarily robust and reliable in wider application) can be expected from NIRS calibrations which were developed using reference samples of pure cultivars grown at

one location, and also from samples with a much narrower range of the substance of interest (DOWELL *et al.*, 2006a).

2. **The reference method by which samples were analysed**. Because the wet-chemistry reference method serves as the foundation for the NIRS calibration, its reliability is in tight positive correlation with the quality of future data that will stem from the NIRS calibration developed on its basis (DELWICHE & GRAYBOSCH, 2002; LU, HUANG & ZHANG, 2006). Calibrations developed from the absolute or normalised amount of each constituent of interest were shown to give better results in comparison with models based on the relative (percent-based) composition of constituents in the sample, as was shown for individual seeds (BAYE, PEARSON & SETTLES, 2006). Thus, the best reference method has to be employed for the NIRS calibration development.

Near infra-red spectroscopy method is sensitive to ambient temperature and humidity, compaction of a sample and its water content, the particle size of the material and reflectance from interfering compounds of the sample. However, these variables can to some extent be reconciled by mathematical data pre-treatments, e.g. 1st and 2nd derivatives, multiplicative signal correction (MSC; sometimes called 'multiple scatter correction') and standard normal variate transformation using statistical software prior to the development of calibration models (MARK, 2001; VAN EYS *et al.*, 2004; FERTIG *et al.*, 2004). NIRS models usually show a bias with a trend to over-estimation of the constituents in samples with the lowest absolute levels and under-estimation in samples with high constituent levels (BAYE, PEARSON & SETTLES, 2006).

The major statistics for the calibration and validation models' evaluation are standard error of calibration (SEC) and the coefficient of determination (R^2) for calibration, standard error of cross-validation (SECV) for cross-validation and standard error of performance (SEP) for the prediction ability (SHENK & WESTERHAUS, 1996; WILLIAMS & NORRIS, 2001). Ideally, the slope of the regression

model would be near 1 and the bias would be near 0. The prediction error is computed as a root mean square error of prediction (RMSEP):

$$\mathrm{RMSEP} = \sqrt{\frac{\sum_{i=1}^{n}(\hat{y}_i - y_i)^2}{n}}$$

where *n* is number of samples (ISAKSSON, MILLER & NÆS, 1992). RMSEP is "the practical average prediction error as estimated by the validation set" (ESBENSEN, 2002), i.e. empirical error estimate given in original measurement units. Double the RMSEP value gives the estimated precision of the model, e.g. the predicted concentration in a new sample is 11% ±0.36%, where 0.36% would be 2×RMSEP (ESBENSEN, 2002). The relative ability of prediction (RAP) index can be calculated (MARTENS & NÆS, 1989) in order to compare the calibration results of different variables:

$$\mathrm{RAP} = \frac{SD_{validation}^2 - SEP^2}{SD_{validation}^2 - SD_{\mathrm{reference}}^2}$$

RAP integrates the SEP value, the variability of the population, and the reproducibility of the analytical reference method, varying from 0 for useless predictors to 1 for perfect predictors (XICCATO *et al.*, 2003). In addition, the value of standard deviation divided by SEP or SECV, called the Relative Predictive Determinant, or the Ratio of Performance to Deviations (RPD), is useful for the evaluation of the precision of an NIRS model (SHENK & WESTERHAUS, 1996; WILLIAMS & SOBERING, 1996; WILLIAMS, 2001; WILLIAMS & NORRIS, 2001; PAULSEN *et al.*, 2003; KOVALENKO, RIPPKE & HURBURGH, 2006). The RPD increases as the standard deviation increases or as the SEP decreases. Values for the RPD range from 1 to 10 with higher values indicating that more variance is explained by the model, hence the calibration model is stronger. Values of 1 or less indicate that the equation predicts about the same results as a random chance. Values in the range of 2–3 are common for starch measurements (PAULSEN & SINGH, 2004).

2.7.4.1. Examples of near infra-red spectroscopy application in grain constituents determination

In research by DOWELL *et al.* (2006a) on wheat NIRS has shown potential in the prediction of protein content, moisture content and flour colour values with accuracies suitable for process control (R^2 >0.97). Many other parameters could be predicted with accuracies suitable for rough screening; however, they are strongly correlated to protein levels. Measurement of protein levels in grain and flour by NIRS is a successful application because protein has very strong and broad absorption bands throughout the NIR region (WILLIAMS, 2001) and it is a major cereal grain component. Stepwise analysis of multiple linear regression models identified eight important wavelengths for protein detection: 1106, 1138, 1156, 1170, 1186, 1200, 1306–1318, and 1500–1504nm (DELWICHE, 1998). The only factors that could be predicted by NIRS with R^2 >0.70 and which were not directly related to protein content were moisture content, test weight, flour colour, free lipids, flour particle size, and the percentage of dark hard and vitreous kernels (DOWELL *et al.*, 2006a). The authors conclude that many traits of grain quality and functionality can be predicted using NIRS, but it is difficult to measure these parameters using NIRS independently of their correlation to protein content.

Some laboratories and NIR instrument producers report accurate reliable results for starch content readings on whole grain, which are obtainable from NIR calibrations on diode array NIR analyser DA 7200 (STAERK & GRUNEWALD, 2006) and NIT instrument FOSS Infratec™ 1241 (FOSS, 2008). Pioneer Hybrid International reported the development of a NIR instrument for accurate point-of-sale measurement of HTF ('high total fermentable') on maize whole grain (BUTZEN, HAEFELE & HILLARD, 2003; BOTHAST & SCHLICHER, 2005). However, SMITH *et al.* (2006) reported NIR calibrations for starch that appeared insufficiently accurate for a reliable estimation of ethanol yield from wheat grain, but calibrations for protein were more robust and thus the authors suggest that the protein calibrations are usable for ethanol yield predictions at the initial stages of research. In addition, it was suggested that it

would be possible to develop NIR calibrations for expected ethanol yield, with direct measurement from grain samples (UTHAYAKUMARAN, BATEY & WRIGLEY, 2005). Some encouraging results were obtained in a GREEN grain project, which showed that it was possible to explain the ~80% variation in the ethanol processing yield by direct NIR prediction, which should prove adequate for use in trade (SYLVESTER-BRADLEY & KINDRED, 2008).

Starch is usually inversely proportional to the protein content in grain and a negative protein peak occurs at about 870–872nm for high-starch samples (PAULSEN & SINGH, 2004). DELWICHE *et al.* (1998) reported $R^2 = 0.99$ for protein content prediction in hard red winter wheat, and MILLAR (2003) reported a similarly excellent result ($R^2 = 0.99$) for protein content measured from whole grain and from flour. Moreover, protein fractions (gliadin and glutenin) were predicted by means of NIR techniques, which may be of use for breeding programmes (DELWICHE *et al.*, 1998; WESLEY *et al.*, 2001a).

Extractable starch is correlated to total starch content in a positive direction, but usually with R^2 values in the range of 0.6 to 0.7 (SINGH *et al.*, 2002). Starch content refers to the amount of starch present, while extractable starch refers to the amount of starch that can actually be extracted (its output or yield). An NIR calibration for maize starch yield was developed from 940 samples, where the starch yield ranged from 58 to 72% (DWB), and SEP of 1.06, R^2 of 0.77 and RPD of 2.1. This indicates that about 95% of similar samples could have starch yield predicted by NIR within about ±2.1% (PAULSEN *et al.*, 2003). NIRS calibration with the ability to predict starch yield with a high correlation coefficient of validation ($R^2 = 0.898$) was developed for corn by CHAWNUA (2000). Another calibration for extractable starch in maize was developed from NIT, based on 2267 samples collected over five crop years. The calibration had $R^2 = 0.79$, SEP = 1.24, slope = 1.08, bias = 0.04 and RPD = 2.15 (PAULSEN & SINGH, 2004). NIR technology can also predict starch damage in wheat flour (MORGAN & WILLIAMS, 1995; MIRALBES, 2004).

NIRS regressions for maize were generated by MLR with $R^2 = 0.89$ for starch, 0.89 for amylose and 0.91 for protein content. Correlation coefficients between laboratory values and calculated NIRS values were 0.95 for starch, 0.94 for amylose and 0.95 for protein. Notably, there was no significant correlation between NIRS predicted values and standard laboratory analysis for moisture content (MULUK, 1996).

NIRS equations were developed by XICCATO *et al.* (2003) using PLSR for starch ($R^2 = 0.90$; SEP = 16g.kg^{-1} dry matter, DM) and crude protein ($R^2 = 0.86$; SEP = 5.6g.kg^{-1} DM). The authors noted that this good result for starch concentration prediction was partly explained by the fact that all starch determinations were performed in the Belgian laboratory (which had secured reference data of a high quality). AUFRÈRE *et al.* (1996) also observed a high $R^2 = 0.94$ and good prediction accuracy SECV = 15g.kg^{-1} DM for starch in compound feeds for swine. KAYS *et al.* (2000) obtained good results for protein in cereals with a SEP between 4.9 and 5.5g.kg^{-1}.

NITS calibrations were developed in maize for starch-amylose content (SAC) and grain-amylose content (GAC) using a set of single and double-mutants (CAMPBELL *et al.*, 1999). The NITS prediction model for SAC (R [*sic*] = 0.96, SEP = 5.1%, RPD = 3.8) was of similar precision to the best GAC model (R [*sic*] = 0.96, SEP = 2.7%, RPD = 3.5).

PLS models were produced from NIRS on whole-grain milled rice that were reasonably accurate for apparent amylose content ($R^2 = 0.89$, SEP = 1.3%) and protein content ($R^2 = 0.97$, SEP = 0.13%) (DELWICHE, MCKENZIE & WEBB, 1996). BAO *et al.* (2001) reported accurate NIRS prediction of apparent amylose content in rice starch with $R^2 = 0.91$ and SEP = 1.39%.

LU, HUANG & ZHANG (2006) developed NIRS calibrations from sweet potato samples for total starch content ($R^2 = 0.86$ and SEP = 1.77%), phosphorus content ($R^2 = 0.89$, SEP = 1.53mg.100g^{-1}), apparent amylose content in starch ($R^2 = 0.90$, SEP = 0.98%) and amylose percentage ($R^2 = 0.91$, SEP = 0.88%) with model

parameters sufficient to allow breeders to screen new breeding lines for these quality characteristics.

2.7.4.2. NIRS application in genetics research

NIR spectroscopy data interpreted by chemometrics can also be used in applied genetics and in plant breeding as a convenient screening and classification tool that helps to expose specific gene expression patterns on the phenotypic level (LU & SHENG, 1990; WATKINS *et al.*, 2001; BAO, CAI & CORKE, 2001; WU & SHI, 2004; GROOS, BERVAS & CHARMET, 2004; JACOBSEN *et al.*, 2005; SISSONS, OSBORNE & SISSONS, 2006; SMAIL, FRITZ & WETZEL, 2006; OSBORNE, 2006). It can be applied on unknown material on its digitalised phenome (e.g. proteome and transcriptome data) as an explorative empirical identification tool without prior hypotheses (MUNCK *et al.*, 1998, 2001, 2004) to identify and reveal broad physical-chemical phenotypic characteristics (e.g. mutations), unexpected effects and patterns (JACOBSEN *et al.*, 2005; RUDI *et al.*, 2006; BAO *et al.*, 2006). It can be especially useful for screening large plant populations (NGONYAMO-MAJEE, 2005; SARATH *et al.*, 2008) on bulk samples and individual kernels in early generations (i.e. F_2 and F_3) for highly heritable traits (BAO, CAI & CORKE, 2001; BAO, SHEN & JIN, 2007).

Near infra-red spectra can be interpreted by PCA, when near infra-red spectra representing the total effects of genetic covariance (pleiotropy and linkage) of the genes of interest are being compared to a preferably near-isogenic background (MUNCK *et al.*, 2004). Environmental differences are mainly expressed as spectra offsets from the baseline, thus for the best genetic separation by NIRS, the plant material under investigation should be grown in the same environment (MUNCK *et al.*, 2001).

Barley flour NIR spectra were used as a tool to differentiate between normal and high lysine barley mutants and these spectra characterise associated large changes in percentage of starch and (1/3,1/4)-β-glucan (DOLL, 1983; MUNCK *et al.*, 2001, 2004), and can also be used for analysis of barley genetic diversity in gene banks

(MUNCK, 2003; MUNCK & MOLLER, 2004). NIT measurements were used for the classification of maize endosperm mutants (CAMPBELL, SYKES & GLOVER, 2000). NIR applied on ground grain of durum wheat showed that waxy kernels had a spectroscopic resemblance to softer wheats (VIGNAUX *et al.*, 2004). VELASCO & MOLLERS (2002) used NIR reflectance spectroscopy as a screening tool in segregating populations of rapeseed (*Brassica napus* L.) for protein determination.

DELWICHE & GRAYBOSCH (2002) conducted a study of using NIRS for waxy wheat identification, and differentiating them from partially waxy and wild-type phenotypes. It was demonstrated that within a crop year, near-perfect separation of fully waxy from non-waxy lines was achievable, but further classification for the correct number of active GBSS genes was more difficult, with an average overall accuracy of 60%.

2.7.4.3. NIRS application for individual kernel sorting and classification

Rapid screening of individual kernels for multiple chemical constituents and selection for desired traits can be realised based on single-seed classification by NIRS/NITS, which is of especial interest in plant breeding because of the non-destructiveness and rapidity of this process (DELWICHE, 1995; DELWICHE & HRUSCHKA, 2000; COGDILL, HURBURGH & RIPPKE, 2004; PASIKATAN & DOWELL, 2004). It has the potential to allow the identification of individual kernels that deviate significantly from the mean composition within a population, and to give an indication of abnormal distribution of kernels within the sample, as a routine homogeneity analysis. It can be applied for the identification of outlying individuals, or for the sorting of kernels with different compositions, taken from a segregating population, in order to increase the purity of heterogeneous lines (NIELSEN, PEDERSEN & MUNCK, 2003; DOWELL *et al.*, 2006b).

Single-kernel NIRS scans were used by DELWICHE & MASSIE (1996) to classify wheat according to a five-class model with an accuracy ranging from 65% for soft red winter wheat to 92% for soft white wheat. SONG, DELWICHE & CHEN (1995)

reported successful classification accuracies (>94%) for artificial neural network models developed from NITS of single kernels.

DELWICHE *et al.* (2006) developed NIRS-based identification of waxy genotypes in single tetraploid (durum) wheat kernels for classification by waxy allele: waxy (double-null), partially waxy (*wx-A1* null, or *wx-B1* null), or wild type, which was often >95% correct for waxy genotypes.

DELWICHE (1995) showed the feasibility of measuring protein content on individual wheat kernels using near infra-red transmittance spectroscopy in the range of 850–1050nm. ABE *et al.* (1996) used the same technique but developed models using combinations of selected wavelengths. Models that used the average of spectra, taken from four different directions relative to the kernel, yielded the lowest standard error of prediction. It demonstrated that spectral averaging could minimise kernel shape effects.

DELWICHE (1998) has developed the NIRS method for protein measurement in single wheat kernels by using reflectance spectra at 1100–1500nm from individual kernels, oriented crease-side-down. The spectra were used to develop calibration models for single wheat classes, for classes pooled according to colour, and for all five USA wheat classes. The researcher obtained a SEP of 0.46–0.72%, which was dependent on the modelling technique.

Calibrations developed for single-kernel sorting of wheat based on their protein content showed $R^2 = 0.92$ and SECV = 0.47% when five PLS factors were used (DOWELL *et al.*, 2006b). In analogous study of DELWICHE & HRUSCHKA (2000) these model parameters were $R^2 = 0.91$ and SECV = 0.37%. In the same publication DOWELL *et al.* (2006b) report that the calibration developed for sorting proso millet (*Panicum miliaceum* L.) according to waxy/non-waxy characteristics resulted in $R^2 = 0.65$ and SECV = 0.29 with six PLS factors. A cross-validation showed that all waxy samples and 89.5% of the wild-type samples were correctly identified.

A commercial colour sorter equipped with near infra-red filters was successfully evaluated by PASIKATAN & DOWELL (2004) for its potential to sort high-

and low-protein single wheat kernels for high-protein class (>12.5%, 12% moisture basis) or low-protein class (<11.5% protein). NIELSEN, PEDERSEN & MUNCK (2003) developed a good and robust calibration model for protein content, based on single seed NIT spectra corrected by the second derivative followed by MSC.

NIR and NIT spectroscopy was applied for individual maize kernels, for the purpose of sorting them for different types of fungal diseases (PEARSON *et al.*, 2001; DOWELL *et al.*, 2002; PEARSON, WICKLOW & PASIKATAN, 2004) and also for the identification of genetically modified seeds (MUNCK *et al.*, 2001). Spectra obtained on kernels with NIR showed to be of better quality compared to NIT spectra, because the latter are more sensitive to the density or total mass of samples, and tend to contain excessive levels of noise because of scattering. Thus, NIR is more suitable for solid samples such as grain (MARK, 2001; COGDILL, HURBURGH & RIPPKE, 2004; BAYE, PEARSON & SETTLES, 2006), and is easier to adapt to real-time analysis than NIT (DELWICHE, 1998; PASIKATAN & DOWELL, 2004).

As can be seen from the above examples, a number of conventional analytical and near infra-red instrumental methods exist for assessing the quality of grain; these methods were shown to be successful in practice. Therefore, near infra-red technology has room for more extensive implementation in plant breeding and genetics applications, in particular for the development of improved specialty starches and in the breeding of cereals for the ethanol industry.

2.8. STARCHY GRAIN AS THE SOURCE MATERIAL FOR BIO-ETHANOL PRODUCTION

2.8.1. Starch fermentation and distillation process

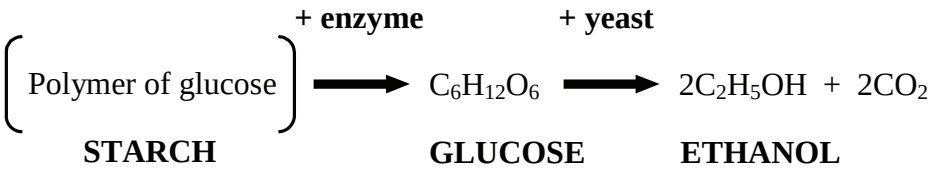
Ethanol yield from the source material depends on the amount of starch and other fermentable sugars in the feedstock, the conversion ratio of starch into fermentable sugars, and the fermentation efficiency of these sugars into ethanol (SMITH *et al.*, 2006). Conversion of starchy grain into ethanol is a complex process

and is a mix of technologies that include microbiology, biochemistry and engineering. The common fermentation and distillation process (dry-grind process) consists of the following steps (after BOTHAST & SCHLICHER, 2005):

1. **Mechanical grinding** of the grain as finely as practical. This step can significantly affect ethanol yield because the difference between fine and coarser ground meal may result in a difference of between 5 and 10% in the ethanol yield (KELSALL & LYONS, 2003).
2. **Slurry preparation** – starchy raw material is mixed with warm water to create mash (slurry); the pH is adjusted to 6.0 and thermostable α-amylase is added; this breaks down the starch polymer into soluble complex sugars by the hydrolysation of α-1,4 bonds. The next two steps (**gelatinisation** and **liquefaction**) are also jointly known as **hydrolysis**.
3. **Gelatinisation** – the mash is heated to above 100°C in a jet cooker (this takes a few minutes), resulting in the cleavage and rupture of starch molecules having a high molecular weight (TESTER, KARKALAS & QI, 2004b). When the mash is heated to about 50°C the amylose in the starch granule swells, the crystalline structure of the amylopectin disintegrates and the granule ruptures; it results in the starch being easily digestible (SAJILATA, SINGHAL & KULKARNI, 2006).
4. **Liquefaction** (for about 30 minutes) – the temperature is lowered to 80–90°C and more α-amylase is added, greatly reducing the size of the starch polymer. The final product of the amylolytic breakdown of starch is maltose (a disaccharide of glucose); its production is accomplished through a chain of intermediate products of hydrolysis, namely dextrins (KRETOVICH, 1986). During the first stages of hydrolysis dextrins are created, not differing much from starch in their molecular size and chemical qualities. Dextrins turn blue or violet when coming into contact with iodine. During consecutive hydrolysis processes, the molecular weight of these dextrins declines, their deoxidising ability increases and

they start to turn dark-brown, and then red in the presence of iodine, and then finally cease to react with iodine. According to their molecular weight, dextrins are divided into amylodextrins, erytrodextrins, achrodextrins and maltodextrins (BONNER & VERNER, 1976).

5. **Saccharification** – the mash is cooled to 65°C (or 32°C depending on the technology), the pH is adjusted to 4.5 and glucoamylase is added, converting the remaining short-chain carbohydrate polymers into glucose.
6. **Fermentation** (40–72 hours at 32°C) – fermentation is equivalent to catalytic burning in which 49% of the input glucose is converted into CO_2 (PATZEK, 2004):



A specially selected industrial strain of yeast (*Saccharomyces cerevisiae* L.) is added to mash, and often also a nitrogen source (in the form of carbamide or ammonium sulphate) to promote the growth of yeast. Proteases can also be added that help to release free amino acids from proteins, serveing as an additional source of nitrogen to yeast (GENENCOR, 2006). Phytic acid was shown to have a significant anti-nutritive effect on yeast because it forms complexes with minerals and amino acids (KELSALL & LYONS, 2003). As a result of CO_2 build-up, the pH falls below 4.0 helping to control the growth of contaminating bacteria. The mash has a final ethanol concentration of 8–12%.

7. **Distillation** – after the fermentation process is completed, the fermented mash (wort) is distilled through the distillation column for ethanol separation by application of heat and use of the difference in boiling temperature of ethanol (78°C) and water (100°C, at sea level). The distillation column produces ethanol with ~95% concentration (azeotrope

with water), and by use of molecular sieves the product can be further purified into absolute (100%) ethanol.

Therefore, the common ethanol production process has three main steps – **gelatinisation** and then **saccharification** to make glucose from starch, followed by **fermentation** to convert the glucose into ethanol. Many modifications to the fermentation process were developed and implemented. Often simultaneous saccharification and fermentation (SSF) is employed because it has advantages such as being generally more energy-efficient, having a lower initial osmotic stress on yeast, a lower possibility of microbial contamination and being able to provide up to 8% higher ethanol yields (BOTHAST & SCHLICHER, 2005). Very-high gravity (VHG) fermentation technology (in contrast to conventional in fuel alcohol manufacturing 'normal gravity' levels of 20–24g of dissolved solids per 100g of mash, i.e. 20–24°Plato) was developed that gives an increased final ethanol concentration and reduced processing costs (INGLEDEW, 1993; THOMAS, HYNES & INGLEDEW, 1996; WANG *et al.*, 1999).

Another major advance in technology is the development of enzymes (such as STARGEN™) that degrade raw, uncooked starch, thereby improving overall process economics (GENENCOR, 2005a; SHETTY, LANTERO & DUNN-COLEMAN, 2005; BHARGAVA *et al.*, 2005; GRAY, ZHAO & EMPTAGE, 2006). What is even more interesting is the design of yeasts that can produce such enzymes and thus can grow on raw, uncooked starch. For instance, Bio-energy Corp. (Osaka, Japan) has developed a one-step process for simultaneous liquefaction, saccharification and fermentation using bio-engineered yeast, which works at 30–38°C and gives an ethanol output* of 92% from the theoretical model (ONDREY, 2005).

Starch hydrolysis requires one molecule of water per molecule of glucose, thus 1000kg of pure starch can be potentially converted into 1111kg of glucose ($C_6H_{12}O_6$).

* In many instances in literature, the terms 'ethanol yield' and 'ethanol output' are used interchangeably. However, it seems to be more appropriate to use the term 'ethanol output' when talking about the volume of the substance obtained from the unit of grain weight (e.g. L.tonne^{-1}), and 'ethanol yield' when talking about its volume as calculated from the unit of field area (e.g. L.ha^{-1}).

This glucose (assuming perfect fermentation efficiency) would be converted into 568kg of ethanol (C_2H_5OH), which is equal to 720L with a density of 0.789kg.L^{-1} (SMITH *et al.*, 2006). Other authors take the theoretical efficiency of ethanol recovery from starch as 51%, thus 1kg of dry cereal grain with 65% starch content (DWB) may yield 0.65 × 0.51 = 0.332kg of anhydrous ethanol with zero losses (SANCHEZ *et al.*, 1988; PATZEK, 2004). Efficiency of the starch hydrolysis and its breakdown to fermentable sugars is about 98–99% considering 1–2% starch content in residue DDGS (distillers' dried grains with solubles) (SMITH *et al.*, 2006). The average empirical conversion rate of starch and glucose into ethanol by fermentation for cereals is about 90–95% (THOMAS, HYNES & INGLEDEW, 1996; LOYCE, RELLIER & MEYNARD, 2002; WU *et al.*, 2006). The imperfect conversion efficiency can be explained by incomplete hydrolysis of starch, glucose consumption by yeast due to its growth during fermentation, and production of by-products (THOMAS, HYNES & INGLEDEW, 1996; KOSARIC & VARDAR-SUKAN, 2001).

According to SMITH *et al.* (2006), ethanol yield (which in this case more correctly has to be called 'ethanol output') varied between 410 and 480L.$tonne^{-1}$ of wheat grain in their research. Thus, the benchmark ethanol yield of average UK feed wheat can be taken as 435L.$tonne^{-1}$ of dry grain at 84% actual efficiency taking 518L as a theoretical yield, with calculations based on DWB having 11.5% protein, 69% starch and 3% sugar. The authors argue (the statement is questionable though) that processing efficiency in their research was probably about 92% of theoretical potential, because yeast growth normally requires about 8% of the sugars available for fermentation. In a recent study by DAVIS-KNIGHT & WEIGHTMAN (2008), triticale gave relatively high average ethanol yields of 436L.$tonne^{-1}$ DM grain at 11.5% grain protein with no addition of industrial enzymes. However, it also showed higher residue viscosity compared to that of wheat. The higher free sugar content of triticale grain was given as an explanation for a higher triticale ethanol yield to starch (EY:starch) ratio: 644L.$tonne^{-1}$, compared to that of wheat 630L.$tonne^{-1}$. Another

practical example of EY:starch conversion ratio was shown to be also 630L.tonne^{-1} for wheat, compared to the theoretical value of 661L.tonne^{-1} (KINDRED *et al.*, 2008).

Different cultivars and environments significantly affect the ethanol yield from grain. Variations of 5% in ethanol yields were observed among 16 sorghum samples, and the effect of location on fermentation was as much as 5% for ethanol yield, which in both cases strongly related to chemical composition and physical properties of grain (ZHAN *et al.*, 2003). WU *et al.* (2006b) observed variations of 22% in ethanol yield and 9.1% in fermentation efficiency among 70 sorghum samples. In research with four triticale cultivars grown over 3 years in 4 locations ethanol yields, with the addition of technical enzymes (Termamyl SC and SAN Extra L), were between 370 and 460L.tonne^{-1} dry matter. The starch content was positively correlated with ethanol yield ($r = 0.396$) and negatively correlated with protein content ($r = –0.327$) (KUCEROVA, 2006).

2.8.2. Parameters for assessment of hydrolysis and fermentation

When analysing grain potential for bio-ethanol production it is important to follow the conventional industrial fermentation and distillation process. The most important analytical methods employed for the analysis of raw materials in this process is the measurement of starch content, determination of fermentable substance (FS) and the auto-amylolytic quotient (AAQ). The FS is defined as the sum of the glucose and maltose contents in the raw material, calculated as starch, which can be determined after the raw material is completely digested and dispersed as well as liquefied and saccharified by the addition of technical enzymes (YOOSIN & SORAPIPATANA, 2007). If the ethanol yield related to the FS is determined, the data is based on the digestibility of the starch to fermentable sugars. Another way to base the data on the digestibility of the starch is to determine the AAQ. The AAQ is defined as the percentage yield of ethanol obtained without the addition of saccharifying enzymes, compared to the ethanol yield with the addition of an ideal combination of technical enzymes (SENN & PIEPER, 2000, 2001). The ethanol yield, for this reason,

can be measured using a small-scale laboratory process that emulates commercial, potable ethanol production (BROSNAN *et al.*, 1999).

2.8.3. Role of technical and endogenous enzymes

Starch cannot be directly metabolised into ethanol by conventional yeast, but must be converted from its polymer form into simple sugars prior to fermentation by yeast. In order to achieve a high bio-ethanol yield, the substrate has to be cooked at a high temperature (above 100°C using a jet cooker) and large amounts of amylolytic enzymes (α-amylase and glucoamylase) must be added, resulting in high costs (SHIGECHI *et al.*, 2004; MOJOVIC *et al.*, 2006). It is possible to produce glucose syrup from some cereals like triticale without the addition of technical enzymes. In such processes, starch is hydrolysed directly from the raw material (e.g. cereal flour in the mash) with the aid of endogenous enzymes (GLATTHAR, HEINISCH & SENN, 2004; VUAUROVIA & PEJIN, 2007). For this kind of production technology, only raw materials with a high endogenous enzyme content are usable, which are recognisable at a falling number below 70 units (ANDE, PIEPER & SENN, 1998). The addition of technical enzymes, when starch is degraded, is common practice in industrial ethanol production due to the resultant reliability, speed, and effectiveness gained. The use of technical enzymes seems to be more important than the changes that breeders can make in enhancement of the AAQ at the expense of increased endogenous enzyme content that leads to a risk of pre-harvesting sprouting. In addition, endogenous enzymes can only be used when no high-temperature step is employed for starch gelatinisation, because these enzymes would be inactivated by the high temperature (SMITH *et al.*, 2006). In recent times, tremendous improvements have been made in industrial enzyme efficiency, resulting in increased bio-ethanol yields and a reduction in process time and costs (MABEE *et al.*, 2006; GRAY, ZHAO & EMPTAGE, 2006).

2.8.4. Role of non-starch polysaccharides in fermentation

Genetic variability exists both between and within cereal species with regard to the structural composition of starch and non-starch polysaccharides (NSP) that may interact with starch and affect its degradation characteristics. The effect of NSP on ethanol yield is twofold: NSP displaces starch in the grain reducing the total quantity of fermentable substance (SYLVESTER-BRADLEY & KINDRED, 2008), and their presence restricts the starch gelatinisation process and subsequently reduces its hydrolysis by α-amylase (TESTER & SOMMERVILLE, 2003; BRENNAN & CLEARY, 2005). Pentosans are able to bind large amounts of water, which results in the formation of gels and the increased viscosity of aqueous solutions. Among these pentosans, arabinoxylans are the main polysaccharides that promote viscosity in wheat (SAULNIER, PENEAU & THIBAULT, 1995; SMITH *et al.*, 2006). In general, high-ethanol yielding cultivars tend to give low residue viscosities, which can be explained by negative correlation between NSP and starch content in whole grain (SMITH *et al.*, 2006). Barley and oats β-glucans have a high viscosity and slimy consistency, and because of that, they can cause wort filtration problems in brewing (HAARD *et al.*, 1999). Cultivars of barley having a soft endosperm and thin cell walls, resulting in rapid cell wall modification during brewing, are the preferred raw material (HOME *et al.*, 2001). The addition of enzymes can play an important role in cereals like rye considering their high amounts of NSP. The addition of xylanase and arabinosidase enzyme mixed into mash helps to eliminate high viscosity problems and improve the filtration performance (HOME *et al.*, 2001; SMITH *et al.*, 2006). Compared to rye, triticale does not contain high amounts of pentosans, thus high viscosity is not such a problem during its fermentation (VUAUROVIA & PEJIN, 2007). Some modern cultivars of triticale give low mash viscosity similar to that of soft wheats (DAVIS-KNIGHT & WEIGHTMAN, 2008).

More research is needed into the interactions of fermentable with non-fermentable constituents (mainly NSP) in grain. The critical question would be whether adding technical enzymes would have the same influence over the complete

genetic variation between cultivars and species. In the case of the answer being 'yes' it probably would not make any sense to start with a specific breeding trait, taking into consideration the NSP level for bio-ethanol production. On the contrary, this could be a new opportunity for producing very high grain-yielding feeding cultivars with a relatively higher NSP and higher viscosity levels (JACOBI & HARTMANN, 2005; WELLIE-STEPHAN, 2005; DAVIS-KNIGHT & WEIGHTMAN, 2008).

2.8.5. Factors that affect processing rate and efficiency

Common factors that affect carbohydrate hydrolysis by enzymes, the processing rate and its efficiency include the starch composition (physical and chemical structure of the starch itself, its protection by protein matrices), the concentration of the substrate, the activity and concentration of enzymes and their inhibitors, and the conditions during hydrolysis *viz*. temperature, amount of time, pH, viscosity, and so on (BURGOS-HERNANDEZ *et al.*, 1999; TESTER, KARKALAS & QI, 2004b; SVIHUS, UHLEN & HARSTAD, 2005; TESTER, QI & KARKALAS, 2006; SMITH *et al.*, 2006; BALAT, BALAT & OZ, 2008). Some studies showed that inhibitors of α-amylase are quite stable even when temperatures are high and there are changes in pH levels (BURGOS-HERNANDEZ *et al.*, 1999). The viscosity of the water extract (VWE) from ground rye was shown to be negatively correlated to the size of starch granules. The VWE was more greatly affected by environmental conditions than by genotype (GONCHARENKO & TIMOSHCHENKO, 2006). For ethanol production, starch granules must have a size and form that requires less energy input for the gelatinisation step, however without a reduction in the grain/starch yield (MORELL & MYERS, 2005). In research with sorghum, it was found that cultivars designed for the bio-ethanol industry need to possess the following characteristics: high starch content combined with high yields, quick liquefaction of starch, low viscosity during liquefaction, a high fermentation speed, and high bioconversion efficiency. Factors that negatively affected the bioconversion efficiency of sorghum grain were phenolic

compounds, tight-storage protein matrix, its low digestibility, high viscosity, and high temperature of gelatinisation (formation of amylose-lipid complexes) (WU *et al.*, 2006b). For starch to be readily digestible by enzymes, it must be amorphous (not crystalline); freely accessible to enzymes (not entrapped in bigger particles); preferably solubilised; not in associations or complexes with other molecules (e.g. amylose-lipid complexes or protein matrix); not chemically modified (TESTER, KARKALAS & QI, 2004b; KOLIATSOU & PALMER, 2004).

2.8.6. Traits of interest for grain into ethanol conversion

When small grain cereals, e.g. wheat or triticale, are considered as a feedstock for ethanol production, the following traits have been shown to be valuable and of top priority: choice of highest yielding cultivars with large, well-filled ('plump') grain, low length to width ratio, low or medium protein content, high starch content, high starch turbidity (easy extractability), low residue viscosity, and no fungal contamination (no mycotoxins, good ear fusariosis tolerance) (TAYLOR & ROSCROW, 1990; ANONYMOUS, 2003; WELLIE-STEPHAN, 2005; SWANSTON *et al.*, 2005, 2007; SMITH *et al.*, 2006; KINDRED *et al.*, 2008). These parameters are similar to breeding objectives for feed wheat of B and C classes (WELLIE-STEPHAN, 2005). Currently low protein content is viewed as the best predictor of high ethanol yield for small grain cereals (DAVIS-KNIGHT & WEIGHTMAN, 2008).

2.8.6.1. Industrial requirements

Traits of interest and specifications of feedstock used for bio-ethanol production may vary from one distiller to another, depending upon the industrial process in use, and the co-products produced. Current industrial specifications for wheat as a feedstock for potable ethanol production in the UK are as follows: soft wheat, with test weight above $0.72 kg.L^{-1}$ and as low a nitrogen content as possible. Other continental European distillers who use the dry-grind process require wheat to be of a standard feed grade, because of its low price, low protein content, and thus

high starch content. Additional requirements include a grain moisture of less than 15%, low mycotoxin levels, no ergot, no heavy metal contamination, and low foreign matter content (ANONYMOUS, 2003). Some Canadian distillers, who employ a wet-grind process, require specified bread wheat cultivars because of their high protein content for the production of gluten as a co-product, and there are not many differences in yields of feed and bread wheats in Canada (SMITH *et al.*, 2006).

The particular traits now being explored for improved ethanol production from cereals include overall starch production per hectare (because yield per area unit influences production efficiency), starch composition (amylose/amylopectin ratio), and its compositional interactions (MCLAREN, 2005; SMITH *et al.*, 2006). Bio-ethanol yield per hectare is demonstrated to be largely a function of grain yield, thus it depends on the agronomic intensity level, which in turn is mainly determined by the level of nitrogen supply (TAYLOR & ROSCROW, 1990). Yield is also determined by other factors such as location, soil type, fertility, previous crop, management, environmental conditions and yield potential of the crop (ROSENBERGER *et al.*, 2000; SMITH *et al.*, 2006; SARATH *et al.*, 2008). Variation in ethanol yield between sites and years is commonly larger than between cultivars (SMITH *et al.*, 2006). The highest ethanol yields from winter wheat, rye and triticale always occurred when propagated at the highest agronomic intensity level, with triticale being the most efficient crop in terms of cost per litre of ethanol generated (ROSENBERGER *et al.*, 2000, 2002). It can be seen from experience in the potable ethanol industry that wheat cultivars differ both in ethanol yields and in the easiness with which they can be processed. It can be concluded that the main ways to successfully use grain for ethanol production in a given environment is through the choice of cultivar and nitrogen management, by avoidance of late application and over-application of nitrogen (LOYCE & MEYNARD, 1997; SMITH *et al.*, 2006). When nitrogen application was optimised for ethanol yield instead of grain yield, considering relative prices of the fertiliser, grain and ethanol, application of 12–22% lower optimum nitrogen amounts were required for crop growth (SYLVESTER-BRADLEY & KINDRED, 2008).

2.8.6.2. Importance of high starch content

The main grain quality parameter for bio-ethanol is that the grain have a high starch content (WELLIE-STEPHAN, 2005). Starch content was demonstrated to be positively correlated with ethanol yield (in wheat and rye each percentage of starch content giving 4.7L of ethanol per tonne of grain) and negatively correlated with the protein content in the grains regardless of the cereal crop species. Triticale in this research yielded less ethanol per unit of starch content and was less responsive to increases in starch content compared to wheat and rye (ROSENBERGER, 2005). However, triticale was shown to be a feedstock with high ethanol production potential by other research studies, producing comparable or higher ethanol yields at a given protein level compared to wheat (AUFHAMMER *et al.*, 1994, 1996; AUFHAMMER, 1998; FLEISCHER & SENN, 2005; DAVIS-KNIGHT & WEIGHTMAN, 2008). This difference in results can be due to different environmental conditions and different cultivars of crops used, as well as different methodology employed for yield components assessment.

Some data seems to indicate that high extraction of ethanol from grain does not necessarily correlate with high starch content, but high starch content would be a good basic condition for maximising the ethanol extraction (JACOBI & HARTMANN, 2005). For instance, LARSON (2004) reported lack of correlation between extractable starch and ethanol yield in the dry-grind ethanol process for maize, with the explanation that the industrial process may make non-extractable starch still fermentable. It was argued that for the dry-grind process high ethanol yield ('high total fermentable,' HTF, taking into account starch and all other fermentable sugars) is a more accurate indicator of grain quality than total starch or extractable starch content, although a high extractable starch trait is considered to be important for the wet-milling process (ANONYMOUS, 2003; BOTHAST & SCHLICHER, 2005). In any case, grain with lower starch levels and a higher moisture content will automatically make it impossible to obtain high ethanol yields. For example, the use of maize with a higher moisture content (14%) and lower starch content (67%) leads to an ethanol

yield of 365L.tonne^{-1} of grain instead of >400L.tonne^{-1} which is possible from maize with a 12% moisture content and 71% starch content (INGLEDEW, 2005).

2.8.6.3. Importance of low protein content

Grain yield and grain protein content are well known to be in an inverse relationship in different cultivars (SIMMONDS, 1995). It was argued that the use of rightly composed cultivar mixtures would give an advantage in terms of more consistent overall grain yield per hectare and thus a higher ethanol yield because of lower average protein content in grain (SWANSTON & NEWTON, 2005; SWANSTON, NEWTON & SMITH, 2006). A strong negative correlation was reported between ethanol yield and grain nitrogen content for wheat and triticale (RIFFKIN *et al.*, 1990; SWANSTON *et al.*, 2007; KINDRED *et al.*, 2008; DAVIS-KNIGHT & WEIGHTMAN, 2008). Considering the above, it can be expected that high grain-yielding (per unit area) cultivars can be expected to give high ethanol processing yields (per tonne of grain) (SMITH *et al.*, 2006). It was found by AUFHAMMER *et al.* (1996) that higher protein content in grain correlates with lower ethanol yield, but the variations in different ethanol yields (per tonne of dry substance) from different batches at equal protein content levels led to inexact statements. Similarly, RIFFKIN *et al.* (1990) could not use starch content alone as an accurate predictor for ethanol yield in their research. It can be explained by differences in NSP content which is the third major grain component after starch and protein.

According to SMITH *et al.* (2006), ethanol yield from wheat decreases by about 7L.tonne^{-1} (per dry tonne) for every 1% increase of protein content in grain. A formula was developed that describes the negative correlation between wheat protein content and ethanol processing output (EO; L.tonne^{-1}, DM basis): **EO = –7.31 × protein + 519**. A similar formula was offered by other researches: **EO = –7.2 × P + 520**, where **P** is protein content calculated as **P = [grain nitrogen, %] × 5.7** (KINDRED *et al.*, 2007). Recently, KINDRED *et al.* (2008) developed another formula that empowered the explanation for the variation of 69.7% in ethanol output in the

research, taking into consideration both protein (P) and starch (S) content: **EO = –4.758 × P + 1.752 × S + 371**. Maximum ethanol yield achieved per unit area was about 3630L.ha^{-1}. When comparing two wheat cultivars there was on average a reduction in ethanol output of 5.69L.tonne^{-1} of grain for each 1% increase of protein content, with –0.56 starch/protein correlation, nitrogen fertiliser having significant effect on ethanol production because of its direct influence on grain protein content. These results are in agreement with ROSENBERGER *et al.* (2000). Among different protein fractions, low gliadins specifically enhance ethanol processing yield in wheat (SYLVESTER-BRADLEY & KINDRED, 2008). Considering the above-mentioned, all efforts should be made to improve the genetics of grains in the context of seeing starch content as the preferable breeding objective as opposed to traditional protein content, which is more necessary for grains used in human and animal nutrition (INGLEDEW, 2005).

Proper quality of protein in kernels is also important, because the material remaining after the fermentation and distillation is dried and sold as a special protein feed called DDGS, with a crude protein content of about 36% in its dry matter (for wheat; NOLTE, 2006). Thus, increased lysine content in cultivars that are determined for ethanol production could be beneficial. In such case, triticale has an advantage over wheat because of its higher lysine content (LASZTITY, 1984; OELKE, OPLINGER & BRINKMAN, 1989; SMITH *et al.*, 2006). Distillers require protein content in the range of 11.6–13.6% for wheat. Excessive protein content is not desirable because it can cause by-products to stick during drying (LOYCE, RELLIER & MEYNARD, 2002).

2.8.6.4. Importance of low grain hardness

The hardness of the grain, which depends mainly on the cultivar genotype, can negatively influence ethanol yield because grain protein content and hardness are in positive correlation to each other (DAVIS-KNIGHT & WEIGHTMAN, 2008). It is also easier to separate the bran from the kernel of soft grain and thus soft cultivars require less energy for milling (ABECASSIS, 1993; LOYCE & MEYNARD, 1997; LOYCE,

RELLIER & MEYNARD, 2002). From a genetic point of view, hardness/softness and mealiness/vitreousness are controlled independently (WEIGHTMAN *et al.*, 2005). In mealy endosperm, starch granules are loosely packed into a protein matrix which provides air spaces within the endosperm, contrary to the steely (vitreous) endosperm with a tightly packed matrix of protein, starch and cell walls (SMITH *et al.*, 2006). Wheat with soft endosperm and cultivars without the 1BL/1RS rye translocation are preferred for potable ethanol production because they are easier to process; however, the use of chemicals and enzymes in technical bio-ethanol production may make this advantage less important (SMITH *et al.*, 2006; DAVIS-KNIGHT & WEIGHTMAN, 2008). The softness of grain also increases starch extractability/turbidity (SWANSTON & SMITH, 2008; FEIZ, MARTIN & GIROUX, 2008). Starch from barley cultivars with a mealy endosperm was shown to be more readily releasable and had a higher extract turbidity than starch from steely grain (KOLIATSOU & PALMER, 2003). Hard wheat is not preferred for distilling because of its higher protein content, the lesser accessibility of starch for enzymes and the higher energy requirement for milling (SMITH *et al.*, 2006). However, in research done by TAYLOR, CRANSTOUN & ROSCROW (1993) wheat grain hardness was not found to have an effect on ethanol yields. In a recent study however, wheat cultivar with soft endosperm was shown to have produced more ethanol per tonne of grain (on average 7.7L more) than the hard endosperm cultivar at an equal protein level (KINDRED *et al.*, 2008). In research with sorghum, the digestion of starch from floury endosperm was significantly higher than from vitreous endosperm, yet there not much of a difference was displayed between floury and vitreous maize. The lower digestibility of vitreous sorghum was explained by a higher content of disulphide bond-cross-linked prolamin proteins and their more extensive polymerisation on being cooked (ZHANG & HAMAKER, 1998; EZEOGU, DUODU & TAYLOR, 2005).

2.8.7. Starch bioavailability – effect of amylose and amylopectin

The bioavailability of starch may differ among cereal cultivars and may affect the conversion rate and final yield of ethanol (MOORTHY, 2002). Besides the total starch content *per se*, it seems possible that a change in the ratio of starch components (i.e. amylose and amylopectin) and granule morphology and size, its extent of crystallinity or damage could also alter kernel processing characteristics and influence ethanol yield from grain (THEMEIER *et al.*, 2005; RUDI *et al.*, 2006; SAJILATA, SINGHAL & KULKARNI, 2006). Factors that reduce bioavailability of raw (non-gelatinised) native starches for enzymatic digestion, among others (see HOOVER & ZHOU, 2003) include: α-amylase inhibitors (FLINTHAM, EVERS & KRATOCHVIL, 1993; BURGOS-HERNANDEZ *et al.*, 1999); increased amounts of B or C type crystallites in starch (GERARD *et al.*, 2001; THEMEIER *et al.*, 2005; SAJILATA, SINGHAL & KULKARNI, 2006); amylose-lipid complexes (SENEVIRATNE & BILIADERIS, 1991; GURAYA, KADAN & CHAMPAGNE, 1997; LAURO *et al.*, 1999; NEBESNY, ROSICKA, & TKACZYK, 2002); increased 'blocklet' size of amylopectin (GALLANT, BOUCHET & BALDWIN, 1997); small granules with high crystallinity and high amylose content (THEMEIER *et al.*, 2005; STEVNEBØ, SAHLSTRØM & SVIHUS, 2006), or the prevalence of large A-type granules (over small B-type granules) with a smaller surface area to volume ratio (BROSNAN *et al.*, 1999; TESTER, KARKALAS & QI, 2004b; SVIHUS, UHLEN & HARSTAD, 2005; TESTER, QI & KARKALAS, 2006; STEVNEBØ, SAHLSTRØM & SVIHUS, 2006). However, high amylose content seems to be the most pronounced and acknowledged factor that causes reduced bioavailability of virtually all starches in different technological conditions. This in turn causes lower ethanol output (LEE, SWANSON & BAIK, 2001; NODA *et al.*, 2002; MANGALIKA *et al.*, 2003; WU *et al.*, 2006; RUDI *et al.*, 2006; HUNG, MAEDA & MORITA, 2006; STEVNEBØ, SAHLSTRØM & SVIHUS, 2006; SHARMA *et al.*, 2007). Resistant starches that are found in residue after amylolytic hydrolysis of liquefied starches has taken place are composed mainly of retrograded (recrystallised) amylose. The content of resistant starch increases as the amylose content in starch increases, i.e. they are in strong positive correlation with

each other (SIEVERT & POMERANZ, 1989; RICHARDSON, JEFFCOAT & SHI, 2000; BRUMOVSKY & THOMPSON, 2001; EVANS & THOMPSON, 2004; THEMEIER *et al.*, 2005; SAJILATA, SINGHAL & KULKARNI, 2006). It can be partly explained by means of the formation of lipid-amylose complexes with monoacyl lipids (LAURO *et al.*, 1999; TESTER, KARKALAS & QI, 2004b; WU *et al.*, 2006).

Amylose content varies considerably in different starches and genetic modifications in cereal and tuberous crops have been produced to create starch with amylose contents varying from zero to >75% (KARADJ, STODDARD & MARSHALL, 1999; MOORTHY, 2002). According to DOMBRINK-KURTZMAN & KNUTSON (1997) in studies with maize, starch granules in soft endosperm generally contain more amylose than those in hard endosperm, which influences the digestible substance yield after grinding. Dent maize was observed to yield the most ethanol and the least fermentation residue when compared to waxy, high-lysine, and white maize, while waxy maize yielded the most fermentation residue of all maize (WU, 1989). EVANS & THOMPSON (2004) conducted research to estimate resistance to α-amylase digestion of native granules of high-amylose maize starch genotypes *viz*. amylose extender (*ae*). These genotypes showed high resistance to α-amylase. The correlation between amylose and resistant starch contents was shown to be strongly linear for maize starches (BROWN *et al.*, 2001). OKUDA *et al.* (2005) obtained similar results for rice – digestibility of rice starch was negatively correlated negatively with the amylose content in it. These findings are in agreement with the results of TESTER, KARKALAS & QI (2004b) who studied amylolytic hydrolysis of waxy, normal, and high-amylose starches.

High-amylose cultivars in all crops are shown to be less productive than cultivars with normal starch (JOBLING, 2004). Considering this, it seems that breeding of high-amylose lines for bio-ethanol production is not feasible. However, it does not preclude breeding of high-amylose lines to be used in other industrial applications (TETLOW, 2006).

Waxy starches have higher crystallinity that makes them more readily damaged by milling and thus more easily hydrolysed by α-amylase (TESTER, 1997a; BETTGE, GIROUX & MORRIS, 2000; MIRALBES, 2004). They give higher ethanol outputs and may require shorter fermentation time periods compared to high-amylose starches (SHARMA *et al.*, 2007). WU *et al.* (2006) have observed that amylose content has a significantly adverse effect on fermentation efficiency; that is, the efficiency of cereal starches to ethanol conversion decreased as the amylose content in starch increased, particularly when the amylose content was above 30%. It was found that waxy wheats were more efficient substrates for ethanol production compared to that of normal wheat, maize of various amylose contents, and waxy and non-waxy sorghum. In addition, high-temperature cooking (≥160°C) was necessary for high-amylose maize to obtain a conversion efficiency equal to normal maize or pure starch. These results are in agreement with SMITH *et al.* (2006), who showed that high-amylose starch is unlikely to be economical because of its high-energy requirements for gelatinisation. Waxy wheat requires lower temperatures (85°C) for its gelatinisation compared to normal or high-amylose wheat and does not 'set back' or retrograde to the same extent on cooling, thus adding yet another advantage to the ethanol processors as lower energy input is required (GRAYBOSCH, GUO & SHELTON, 2000; SMITH *et al.*, 2006; SARATH *et al.*, 2008).

According to SMITH *et al.* (2006), waxy wheat grain yields are not comparable to current feed wheat yields, thus hindering their usage for ethanol production. In other studies of waxy wheat, the total starch content was lower compared to wheat with wild-type starch having a concurrent higher level of NSP (i.e. arabinoxylans and β-glucan) with a resultant lower flour yield (YASUI, SASAKI & MATSUKI, 1999; TAKATA *et al.*, 2007). Waxy varieties of barley are also lower in total starch than non-waxy varieties (XU *et al.*, 1997). However, results of other studies with wheat under field conditions showed that introgression of the null *wx* alleles did not result in statistically detectable grain yield reduction (MIURA *et al.*, 2002).

In a study with rice when high-saccharifying fungi and high-ethanol-tolerant yeast were used, waxy and low-amylose (12–20%) cultivars showed the highest rice wine yield with the highest ethanol recovery, followed by intermediate (20–25%) and high-amylose (>25%) rice. Contents of amylose and protein ($r = –0.68$, $P = 0.05$), as well as ethanol recovery and amylose content based on either dry matter ($r = –0.86$, $P = 0.01$) or starch content ($r = –0.85$, $P = 0.01$) were negatively correlated. Starch content in the residual mash was positively correlated with amylose content ($r = 0.88$, $P = 0.01$) (SANCHEZ *et al.*, 1988). However, the results of the study are hardly comparable to others because no conventional fermentation process was followed.

Considering the reviewed literature sources, there is some information available regarding the effects of amylose content in starches and grains on ethanol production efficiency, with a general trend exposed. However, no waxy cultivars of wheat that are bred for every climatic zone are readily available, and no waxy triticale cultivars exist to date, which makes it difficult to extrapolate their possible starch yield and ethanol conversion performances in a given environment. Because of this, little data is available on the influence of the amylose/amylopectin ratio and other yield components of crops on ethanol yield per area unit that, ultimately, directly influences interests of raw material producers (farmers), as well as the bio-ethanol distillation industry. Therefore, information on the desirable ratio of amylose to amylopectin for each cereal crop in a given environment should be of interest to breeders who are aiming at developing new high-ethanol yielding cultivars.

2.9. BREEDING OF CEREALS FOR BIO-ETHANOL PRODUCTION

> "A tasty starch-based recipe for the future is to take a genome sequence, diversity-generating tools, some smart high-throughput phenotyping systems, a few educated guesses on target genes, and shake vigorously. Some exciting science will certainly result, and given the importance of starch […], there is every prospect of there being a few good outcomes…" (MORELL & MYERS, 2005)

2.9.1. Characteristics of an ideal bio-fuel crop

An ideal bio-fuel crop should have a sustained capacity to capture and convert the available solar energy into harvestable biomass with maximal efficiency and with minimal inputs and environmental impacts. A broad description of the properties of an 'ideal' bio-fuel crop is described below (after HEATON *et al.*, 2004).

Maximum efficiency of light use. The economic yields and energy efficiency are determined predominantly by the amount of fermentable substance that can be formed per unit area and per unit of investment of other resources, notably nitrogen. The potential limit on FS yield would be set by the amount of light available, its efficiency of interception, and the efficiency with which intercepted light is converted into starchy grain mass (JIANG, TAO & CAO, 2002; PAN, ZHU & CAO, 2007).

Water content and water use efficiency. Ideally, the harvested grain should be as dry as possible. High moisture content will require an input of energy for drying. Water use efficiency is another important criterion in selecting fuel crops. Available soil water is a significant limitation to crop production on much arable land, and irrigation requires significant inputs of energy whilst placing a demand on diminishing water resources.

Nitrogen and other nutrient use efficiency. Nitrogen use efficiency is determined at three levels; maximisation of them would lead to a higher efficiency of nitrogen use. Firstly, the *efficiency of capture (recovery)* of nutrients from the soil (the amount of nitrogen taken up per unit of nitrogen available in the soil). Secondly, the *efficiency of energy transduction* into biomass in photosynthesis per unit of nitrogen

invested in the photosynthetic apparatus. Thirdly, the amount of nitrogen, and other nutrients, translocated out of the canopy components on their senescence, either into other leaves (temporary storage) or storage organs, i.e. its *efficient internal utilisation* and recycling, in other words nitrogen 'sink' strength (the amount of grain formed per unit of nitrogen taken up from the soil).

Cultivation and control of diseases and pests. Cultivation operations, which include ploughing, planting, and chemical applications, all constitute energy inputs. Fuel crops need therefore to have a life cycle that would minimise the need for these operations. Selection of crops with minimum or nil requirements for fossil-derived fertilisers, pesticide, fungicide, and herbicide applications would help energy efficiency and the environmental acceptability of the bio-fuel production. Selection of non-food crop species and maintenance of genetic diversity is likely to minimise losses due to pests and diseases. Selection of plant species that occur naturally in monotypic stands may also be advantageous.

Minimised changes in land use and farm machinery. Energy crop acceptability would be greatest, and costs of conversion least, if species/cultivars selected as fuel crops could be:

1. planted and harvested with the machinery used for food crops;
2. easily eradicated should the landowner subsequently want to change land use;
3. providing harvestable material in a short period of time.

Environmental impacts and benefits. Some energy crops could have added environmental benefits over current food crops. Perennials that provide above ground structures throughout the year may offer wildlife refuges. The production and turnover of belowground storage organs would add organic matter and carbon to the soil. In comparison to annual plants, perennial plants have more extensive root system, which is in the soil throughout the year. This provides increased resistance to soil erosion and a more effective means for the trapping of nutrients and prevention of nitrogen loss to drainage waters. Because the crop is not used for food, the land could

also be suitable for the spreading of sewage sludge and farm effluents that may represent health risks in areas sown with food crops.

End uses. Alternative markets would be important to maintain price stability. The ideal crop would provide biomass suitable as a feedstock for a range of bio-based industrial processes – such as fermentation, bio-composites and paper production.

More specific properties for the ideal source of biomass for bio-ethanol production include (LOYCE & MEYNARD, 1997; HEATON *et al.*, 2004; SYLVESTER-BRADLEY & KINDRED, 2008):

- **high yield** – thereby reduced land requirements; it is a derivative of crops having a high resistance to disease and pests;
- **relatively high proportion of desirable yield component and tissue** – e.g. a high yield of grain with a high starch content;
- **good quality** – suitable chemical composition for efficient processing, i.e. a high content of fermentable substance (starch and sugars), minimal protein, and low NSP, oil and ash;
- **good agronomic performance** – little or no demand for nitrogen application, efficiency of nitrogen use, minimal cultivation requirements, sufficiently dry biomass at time of harvest;
- **large straw yield** – organic matter that can be re-introduced into the soil.

Some of these characteristics are mutually exclusive. Because different genetic mechanisms influence ethanol yield, it is possible to develop improved breeding lines by combining desired ethanol yield traits from complementary parents (SWANSTON *et al.*, 2007).

2.9.1.1. Breeding for efficient nitrogen use

When sustainability of bio-ethanol production and reduction in emissions is taken into consideration, reduction in input of nitrogen (as well as all other fossil-based material and energy inputs) is much more important than increase in ethanol

output per tonne of grain (DAVIS-KNIGHT & WEIGHTMAN, 2008). The average efficiency of nitrogen recovery by wheat from the soil is 60% and it is possible to improve the trait by means of breeding (BLOOM *et al.*, 1998; FOULKES, SYLVESTER-BRADLEY & SCOTT, 1998; KINDRED *et al.*, 2007). The efficiency of energy transduction into biomass per unit of nitrogen invested in the photosynthetic apparatus involves the root system, its depth and density, its interaction with soil processes, aiding the minimisation of both the quantities of nitrogen that needs to be applied as a fertiliser, and the amount of nitrogen lost through drainage. It also encounters nitrogen that is needed for canopy formation and survival, the volume of which can be reduced without affecting photosynthesis (WHITT *et al.*, 2002). There is a need to breed cereals with low nitrogen requirements that would be used for ethanol production (LOYCE & MEYNARD, 1997; SYLVESTER-BRADLEY, 2007). Field trials with zero and a range of other low and optimum levels of applied nitrogen are necessary, in which several cultivars are tested. It would show the most efficient cultivars that only require minimum nitrogen application. Cultivars with low nitrogen requirements can also be determined in two-level nitrogen trials where they would give a low response to nitrogen (SYLVESTER-BRADLEY & KINDRED, 2008). Some triticale cultivars were shown to perform well at reduced nitrogen levels, because of their inherently lower nitrogen requirements (AUFHAMMER *et al.*, 1996; ANONYMOUS, 2000; DAVIS-KNIGHT & WEIGHTMAN, 2008).

Among grain protein fractions, gliadins constitute on average 40% of the protein content in wheat (varying between 37–50% in commercial cultivars at the same nitrogen level). Gliadins are the most responsive to the level of nitrogen supply and thus have significant potential for direct selection for improved nitrogen efficiency and improved ethanol yield (KINDRED *et al.*, 2008; SYLVESTER-BRADLEY & KINDRED, 2008). They have minimal nutritional value because of very low levels of lysine and other essential amino acids, thus not being of much value for DDGS as a co-product. The gliadins level can be safely reduced by breeding, which would give a proportional rise in starch content and would reduce the nitrogen demand by 30%

(WHITT *et al.*, 2002). Size-exclusion chromatography demonstrated that the gliadin content in wheat grain increases approximately by 0.56g per one gram of increase in total grain protein, thus selection against gliadins content could be a feasible approach for general protein level reduction, which in turn could result in increased ethanol yields (KINDRED *et al.*, 2008).

Substantial improvements in nitrogen capture can be made through the utilisation of the introgression of relevant genes from a wild perennial grass *Leymus racemosus* that has a capacity to produce root exudates-inhibitors of ammonium to nitrate conversion in soil. It could help to reduce nitrogen leaching and its denitrification losses, which is especially pronounced in anaerobic soils or with ammonium fertiliser nutrition (SUBBARAO *et al.*, 2007). Another improvement in nitrogen assimilation in plants can be introduced through alteration of the amino acid metabolism via enhancement of activity of the enzyme alanine aminotransferase (GOOD, SHRAWAT & MUENCH, 2004; LEA & AZEVEDO, 2007). In oil rapeseed, introgression of such traits via genetic modification (GM) led to a 50% reduction of nitrogen requirement (GOOD *et al.*, 2007) and could also be applied in cereals like rice, maize and wheat (ARCADIA, 2007; ETTER, 2007; ALDHOUS, 2008). Over-expression of genes that control glutamine synthase and glutamate dehydrogenase was shown to increase plant biomass because of a more efficient nitrogen metabolism (MIFLIN & HABASH, 2002; JING *et al.*, 2004; GOOD, SHRAWAT & MUENCH, 2004).

2.9.1.2. Breeding for efficient light use

Average efficiency of light interception by plants is less than 2% (RAGAUSKAS *et al.*, 2006). There are some successful GM technology applications in enhancement of inorganic CO_2 capture through regulation of ribulose-1,5-bisphosphate carboxylase-oxygenase (RuBisCO) carboxylation enzyme that leads to the improvement of photosynthesis efficiency (SCHWENDER *et al.*, 2004; CAMP, 2005). The introduction of one C4-cycle photosynthetic enzyme in C3 plants was attempted in rice, potato and tobacco, which did not result in an improvement of photoassimilate

production (MATSUOKA *et al.*, 2001; HAUSLER *et al.*, 2002). However, a positive result was obtained when two C4-cycle enzyme genes with their promoters were transferred from maize to rice and were simultaneously expressed, which resulted in a 35% higher photosynthetic capacity and a 22% higher grain yield (KU *et al.*, 2001).

2.9.2. Alteration of cereal carbohydrate complex by breeding for bio-ethanol production

The current use of starchy crops for bio-ethanol production is heavily focused on the development of complex conversion technologies that typically involve a fermentation step. Breeders have to decide now to start breeding programmes for cultivars which are better adapted to the demands of ethanol production. The basic questions for the plant breeder are the dimensions of the expected market for the new cultivar; the exact character of the breeding trait 'suitability for production of bio-ethanol'; which crops will be finally used for the production of bio-ethanol; is there an association with other breeding traits (e.g. the industrial production of starch). Only traits that are constant over the years are of interest and should be focused on.

To be as effective as possible in plant breeding, it is very important to have exact information about what characteristics are necessary for a crop cultivar to be used for the production of bio-ethanol. Breeders need information from the industry in order to create the best possibly adapted raw material for industrial purposes. The non-ruminant feed and distilling markets both require grain with high starch content rather than high protein content, and the emerging bio-ethanol industry has similar demands. For the production of bio-ethanol the distilling industry needs wheat with a high starch content, a low protein content and very low levels of mycotoxins (NOLTE, 2006). A possible profile of a cereal cultivar designed for ethanol production can include such essential characteristics as high grain yield with high starch content, good kernel constitution (specific test weight only), low kernel hardness, kernel healthiness, high intrinsic enzyme activity (with combined sprouting resistance), high

extraction of ethanol (JACOBI & HARTMANN, 2005). A high yield of ethanol should be the most important trait.

DEFRA (2004) research aimed to explore the potential to develop wheat cultivars with an enhanced value for distilling (both bio-ethanol and potable alcohol production) and non-ruminant feeding. Aims of the research were to identify genes and processes that give rise to high starch grains with high ethanol yields, improved amino acid balance, reduced gliadin proteins and reduced input requirements, especially of nitrogen fertilisers. It appears feasible to combine these different attributes in one wheat cultivar because:

1. the bio-ethanol and livestock feeding industries both regard wheat primarily as an energy source, thus their principal requirements are similar;
2. it is likely that an exploitable variation for the individual traits exists in the gene pool, given the absence of past selection for such traits;
3. when gliadins are minimised by breeding, grain starch would increase proportionally and nitrogen demand would be reduced by 30%;
4. considerable 'inactive' nitrogen is contained in true stems. When used as a breeding target, low stem nitrogen should reduce canopy nitrogen content by 30%, without affecting photosynthesis;
5. a 30% reduction in crop nitrogen uptake should result in a 50% reduction in fertiliser nitrogen demand (DEFRA, 2004).

The application of new production technologies, conventional plant breeding, and biotechnological modifications have resulted in significant yield increases (at the same level of inputs) in maize. The major maize seed companies have screened their germplasm for hybrids that produce a higher ethanol yield in the dry-mill process (BRYAN, 2002; MONSANTO, 2007). The results indicate that genetic components for higher ethanol yield do exist, but these have never been specifically targeted in the past. Major crop plants have been bred primarily for food or feed production, and

never had selection pressure to optimise them for industrial ethanol production. Thus, it would seem there is an opportunity to optimise plant use in bio-energy strategies.

It is known that fractional composition and technological properties of the majority of starches significantly depend on soil-climatic growth conditions (FERGASON & ZUBER, 1962; FERGASON & ZUBER, 1965; HIZUKURI, 1969; SHI, SEIB & BERNARDIN, 1994; MYLLÄRINEN *et al.*, 1998; DEBON *et al.*, 1998; TESTER & KARKALAS, 2001). However, one cannot maintain that it is economically viable to ensure high technological properties of starches by growing its sources in areas with the most favourable soils and climate. It seems more expedient to solve the problem through the genetic improvement of starch qualities, first in crops with the biggest useful genetic variability, which can be effectively used in plant breeding (SHANNON & GARWOOD, 1984). Limited diversity in starch and perhaps other critical pathways may preclude current breeding practices from reaching their full potential. Useful variation, especially for grain quality, needs to be generated for these pathways. Perhaps the most efficient way to introduce potentially useful diversity into cultivated crops is to introgress or to transform the abundant allelic variation that is present in other domesticated and wild relatives for selected genomic regions or genes. This approach could provide the allelic variation necessary to further increase yield and provide a much wider range of kernel qualities (WHITT *et al.*, 2002).

From compositional studies SOSULSKI & TARASOFF (1997) concluded that the relative crop ranking for potential ethanol production in an ethanol plant would be, from best to worst: hard red winter wheat; Canada Prairie Spring (CPS) and Soft White Spring (SWS) wheat; durum wheat, spring triticale and winter triticale and hulless barley; CWRS wheat and fall rye. In some processing factories, poor gluten properties of triticale have led to ‘stickiness’ in the extraction processes. While Canadian triticale breeders think this should not be a problem in modern cultivars, the subject has not been researched at the plant-scale level. So far, results regarding ethanol output, ethanol yield and AAQ are only available for a small number of

triticale cultivars. They do not allow estimation of genotypic variation, which is a prerequisite for breeding when improving ethanol production.

2.9.3. Triticale as a source for bio-ethanol production: agronomic characteristics and breeding objectives

Any grain for industrial energy use (e.g. conversion to ethanol) requires:

1. grain yield and price competitiveness in comparison with other grains as a major factor (SWANSTON, NEWTON & SMITH, 2006);
2. plump kernels with a low percentage of thin kernels;
3. high starch content and high conversion rates to ethanol;
4. a market for co-products;
5. a regularised grain supply chain;
6. sufficient tax or other incentives for the ethanol to be competitive with gasoline in the fuel market.

Of these, the first five criteria are readily met by triticale with present conditions in Canada (GOVERNMENT OF ALBERTA, 2006).

The possibility of triticale grain usage as a perspective feedstock for the fermentation industry was considered as far back as in the mid-1980s (FORTUNA *et al.*, 1985). Triticale may play a significant role in the future as a raw material for industrial uses. Its biomass or straw has been considered as a feedstock in ethanol production (MCLEOD *et al.*, 1998). For the production of biogas, triticale could also find an application as a source of renewable energy (PLOCHL *et al.*, 2003). In addition, the autoamylolitic system makes triticale more suitable for ethanol production than wheat if industrial enzymes are not used (SENN, 2000).

Because of its low input requirement, triticale could play a much greater role in environmentally friendly production. Comparative trials of various crops with different triticale cultivars demonstrated that the biological value of triticale cultivars is comparable to the most suitable wheat cultivars for ethanol processing (MCLEOD *et al.*, 1997). It has advantages in terms of a lower nitrogen input requirement, better

performance in light soils and in a 2nd/3rd cereal rotation position compared to wheat (DAVIS-KNIGHT & WEIGHTMAN, 2008). Its grain yields are also similar or better than for other cereals. Canadian triticale cultivars generally have a lower fibre content than wheat, and comparable starch content, fermentable sugars, pentosans, potential ethanol yields, and lower protein content (MCLEOD *et al.*, 1997).

SCHÄFER *et al.* (1997) studied two cereal cultivars, Alamo (winter triticale) and Contra (winter wheat), at two different locations. Two production intensities were compared. Investigations of ethanol production efficiency were made with and without the addition of technical enzymes. For both locations, the higher production intensities led to a higher energy yield per hectare. Nevertheless, the relationship between output and input was much better when the cereals were produced with a lower intensity level (Table 2.9.3.1; SCHÄFER *et al.*, 1997). However, this can be explained by the fact that there were droughts during the year in which the investigations were made, which prevented higher yields. It is not possible to say that lower production intensities always produce higher output/input relations because on one hand, the high-energy input was created by the additional use of a mineral fertiliser, and on the other hand higher production intensities normally result in higher crop yields. Another effect that could be shown is that the addition of technical enzymes was not needed to produce high output/input relations when Alamo or Contra was cultivated. This is possible because the two cultivars have high autoamylolitic enzyme activities.

Table 2.9.3.1. Output/input energy relationship of the ethanol production (SCHÄFER *et al.*, 1997)

Parameter	Alamo (winter triticale)				Contra (winter wheat)			
Production intensity	low		high		low		high	
Addition of technical enzymes	no	yes	no	yes	no	yes	no	yes
Output / Input (loc.: Ihinger Hof)	5.27	5.27	4.78	4.78	4.67	5.03	4.03	4.39
Output / Input (loc.: Oberer Lindenhof)	4.62	4.62	3.94	3.95	3.39	3.78	3.40	3.71

In a study by ROSENBERGER (2005) in Germany, peak starch contents that amounted up to 72.5% (DWB) were measured in wheat samples, followed by triticale and rye. Wheat samples also revealed the largest starch content variability, compared to triticale and rye cultivars. The highest ethanol yield of up to 465L.tonne^{-1} of grain dry matter, as well as the widest yield range was determined for wheat samples. Ethanol yield was affected both by the grain starch content and by its conversion rate, which is the result of intrinsic starch quality affected by the cereal species and the cultivar genetics.

2.9.3.1. Triticale auto-amylolytic activity and technical enzymes involvement

THIEMT *et al.* (2006) conducted a study to estimate quantitative-genetic parameters for ethanol content, ethanol yield and autoamylolytic quotient (AAQ) in a larger set of genotypes and to analyse their associations with yield and other agronomic traits in various environments. Thirty winter triticale genotypes that represent elite breeding material were investigated. Results revealed a significant genetic variation for ethanol content and ethanol yield with and without the addition of technical enzymes. Ethanol content without addition of enzymes was influenced by the nitrogen level, whereas the addition of enzymes showed no differences between the three nitrogen levels.

Various winter triticale and winter wheat cultivars and mixtures of cultivars were tested by AUFHAMMER (1998) for their autoamylolytic activity and ethanol yield. The triticale cultivar with the highest AAQ was Alamo (AAQ of 96.50–96.96%); Lasko only reached AAQ of 87.5–88.1%. Winter wheat cultivar Adular (AAQ of 93.4–93.7%) was better than Contra (AAQ of 87.9–73.9%). The addition of exogenous enzymes on average led to an increase in ethanol yield only of 15L.tonne^{-1} of grain. The AAQ of cultivar mixtures was as good as or slightly better than the average of the pure cultivars. The best results without the addition of exogenous enzymes were obtained by using mixtures with the enzyme-rich cultivar Alamo. Higher production intensity led to a reduction in ethanol recovery by 8–12L.tonne^{-1}

and in a reduction of the AAQ value by 1–2%. In both cases, with and without the addition of exogenous enzymes, ethanol exploitation increased when grain was harvested five weeks later than at the stage of full ripeness. A late harvest date led to pronounced decreases in ethanol yields for those cultivars or cultivar mixtures that were susceptible to grain loss. Postponement of the harvest date led to an increase in the AAQ from 73.9 to 97.5% for Contra. For Alamo, however, only a slight increase was noted (from 96.9 to 98.0%), and because of its high grain yield it had the best ethanol yield of 2360 and 2930L.ha^{-1} at two different locations. The addition of exogenous enzymes to this cultivar increased the ethanol yield by 85L, independent of the location. A good ethanol yield from wheat Contra (i.e. comparable to that of triticale) was only obtained when exogenous enzymes were added, which increased the ethanol yield by 939L.ha^{-1}.

2.9.4. Mutations linked to starch quality and quantity characteristics and their exploitation by breeding

If feedstock quality can be genetically improved, the economics and efficiency of the feedstock-to-ethanol conversion processes could be significantly enhanced. Improvement of an agricultural feedstock for enhanced end use characteristics via genetic modification requires knowledge of the desired quality attributes, the relative economic value of the quality parameters in relation to yield, and genetic variation for the desired traits. For molecular breeding, the additional knowledge of which genes to suppress or add is needed, as well as knowledge of any associated negative consequences of genetic manipulation (VOGEL & JUNG, 2001; JOBLING, 2004). Conventional approaches to starch modification are usually targeted at the elimination of the activity of one individual isoform or a set of enzymes (MORELL *et al.*, 2004). Yet another approach is based on alteration of these enzyme regulatory systems and/or disruption/altering of interactions/coordination between individual enzymes (YU, 2003; MORELL & MYERS, 2005).

Mutations of interest can be screened directly at the phenotype level, which requires a rapid screening strategy. It has the advantage that it does not require an explanation of the underlying cause, thus the approach is open for the detection of new factors and genes that could influence the property of interest. However, phenotypic screening is often time-consuming and thus does not permit the high throughput screening. For the polyploid species like wheat and triticale, phenotype-based screening cannot be expected to be successful because of the buffering effect of multiple genomes (MORELL & MYERS, 2005). Mutations of interest must be combined in their three genomes, if these mutations are not dominant. Thus, biotechnological (GM) methods could be particularly useful in polyploids. Taking another approach, one can screen the known genes that control the property of interest by using PCR-based molecular screening techniques. Use of such approach on wheat genomes provided a strategy for the identification of genome-specific sets of primers for starch-biosynthetic enzymes (BLAKE *et al.*, 2004). A comparable genetics approach between wheat and rice homologous chromosomes and individual genes that control starch synthesis was recently used (LI *et al.*, 2004). In all cases, genetic diversity and variability in populations under investigation is required for the possibility of application of selection pressure (RAHMAN *et al.*, 2007).

Chemical mutagenesis could be employed to create a source of genetic diversity. The effect of mutagens such as ethyl methanesulfonate (EMS) and azide is one that generally leads to an alteration of a few DNA nucleotide bases or their deletion. A high-resolution melting can identify these alterations or deletions (GRAHAM *et al.*, 2005; RAHMAN *et al.*, 2007). Single nucleotide alterations can be screened for by TILLING (targeting induced local lesions in genomes) – a reverse genetic, non-transgenic method that requires production of hybrids between the parental genotype and the germplasm under investigation with consequent mismatches in genome sequence detected by application of single strand specific nucleases and high resolution electrophoresis (COMAI & HENIKOFF, 2006). TILLING was used to produce waxy wheat by the detection of waxy mutations in each of the

wheat genomes and their consequent combination (SLADE *et al.*, 2005). Individual disruptions in targeted genes can be created by using T-DNA or Tos-17 insertions. The technique is not GM because the Tos-17 is an endogenous mobile element (RAHMAN *et al.*, 2007).

Instead of screening the breeding material for naturally occurring or induced mutations, the GM approach such as RNA interference (RNAi) technology could be possibly used to produce alterations in a genome, which has a significant advantage because it can be limited to selected tissues (WESLEY *et al.*, 2001b; RAHMAN *et al.*, 2007). The approach was implemented to create very-high-amylose wheat lines through alteration of its starch biosynthesis (REGINA *et al.*, 2006). Specific genes can be targeted by this technique using micro-RNA (RAHMAN *et al.*, 2007). Other examples of genetic transformation in wheat and other plants were demonstrated using microprojectile bombardment (VASIL *et al.*, 1992; NEHRA *et al.*, 1994) and anti-sense techniques (VISSER *et al.*, 1991; MURRAY & CROCKETT, 1992; MULLER-ROBER, SONNEWALD & WILLMITZER, 1992; SHIMADA *et al.*, 1993). Transgenic rice was developed, which expresses a heat-resistant starch-degrading amylopullulanase that allows the complete breakdown of the starch within hours of processing at a very little cost (YU, 2003).

2.9.4.1. Alteration of starch quality

Starch quality modifying mutations that are responsible for amylose and amylopectin synthesis are relatively easily detectable and employable in new cultivar development in true diploid (2n = 2x) species like rice, maize and barley. This is not the case with genomic allopolyploid species such as bread wheat (2n = 6x; AABBDD) and triticale (2n = 6x; AABBRR) that possess allohexaploid genomes (BÅGA *et al.*, 1999b). Their breeding requires a different strategy for the combination of recessive starch gene mutations from all three sub-genomes in one genotype in order to obtain a cultivar with noticeably different starch qualities, e.g. combination through the conventional breeding of the three null alleles of GBSS-I in one breeding

line to create a genotype that would produce a truly 'waxy' starch type. Successful results following the application of this approach through backcrossing of individual null *wx* wheats were achieved (NAKAMURA *et al.*, 1995; GRAYBOSCH *et al.*, 2003; HUNG *et al.*, 2008). A similar approach could be implemented in triticale through the creation of primary triple-null waxy lines by the combination of *wx* genes from A and B genomes of durum wheat and the R genome from rye. However, waxy mutants in rye are not described to date (Dr FREDERICK STODDARD, Department of Applied Biology, University of Helsinki, Finland, 2008, personal communication; Dr ROBERT GRAYBOSCH, USDA-ARS, University of Nebraska, Lincoln, NE, USA, 2008, personal communication). It is possible to obtain waxy rye through the extensive screening of rye populations for natural mutations (Dr GRANT MCLEOD, Agriculture and Agri-Food Canada, Indian Head, SK, Canada, 2008, personal communication). The method can be expected to be especially fruitful when conventional rye populations are crossed with a self-compatible type with subsequent self-pollination for a few generations and the screening of an obtained large population to reveal hidden recessive genes. Another approach could involve mutagenesis to trigger waxy mutation in rye with subsequent screening of the M_2 generation of a mutagenised population for waxy individuals (Dr FREDERICK STODDARD, Department of Applied Biology, University of Helsinki, Finland, 2008, personal communication).

Yet another method could involve the elimination of the remaining dominant *Wx* protein genes in the single or double null *wx* plants through mutagenesis (KIRIBUCHI-OTOBE *et al.*, 1997; YASUI *et al.*, 1997). It could also be done in triticale through crossing it with waxy wheat to transfer null *wx* genes from A and B genomes, with subsequent EMS mutagenesis of the resulting double null triticale to generate the *wx* mutation in the R genome for triple-null triticale (Dr ROBERT GRAYBOSCH, USDA-ARS, University of Nebraska, Lincoln, NE, USA, 2008, personal communication).

In order to create a high-amylose triticale, it would probably be necessary to combine the recessive forms of two genes, SBEIIb and either SBEIIa or SBEI, with

the consequent screening of the population for these genes using DNA markers (REGINA *et al.*, 2006; RAHMAN *et al.*, 2007; Dr FREDERICK STODDARD, Department of Applied Biology, University of Helsinki, Finland, 2008, personal communication).

Besides the above-mentioned genes with resultant major effects, other minor genes (QTL) with subtle effects also play a role in determining cereal carbohydrate content and qualities; they must not be underestimated in breeding (MOHAMMADKHANI, STODDARD & MARSHALL, 1999a; TETLOW, MORELL & EMES, 2004; TETLOW *et al.*, 2004; YAO, THOMPSON & GUILTINAN, 2004; MORELL & MYERS, 2005).

Because high-amylopectin and high-amylose starches distinctively differ from each other by their physicochemical and technological qualities and areas of application (WHITE, 1994; BURRELL, 2003; SAHLSTRÖM, BÆVRE & GRAYBOSCH, 2006), the creation of cultivars with starches of each type is of interest as independent directions in crop breeding.

2.9.4.2. Alteration of starch quantity

Plant productivity is determined by both the duration and rate of photosynthesis in 'source' organs (i.e. leaves), and the capacity of developing assimilative ('sink') tissues and organs in which to store fixed carbon (WOODROW & BERRY, 1988; ROWLAND-BAMFORD *et al.*, 1990; CHEN & SUNG, 1994; CARRARI *et al.*, 2003; JENNER, 2003; SCHWENDER, OHLROGGE & SHACHAR-HILL, 2004). Yield components of plants can be manipulated through direct genetic transformation by modification of the key carbohydrate metabolism enzyme activity, by increasing or decreasing the 'sink' of metabolites (GEIGER, KOCH & SHIEH, 1996). Higher starch yields can be achieved by targeting AGPase, which is the key enzyme in starch biosynthesis that catalyses the rate-limiting step and produces the glucosyl precursor, controlling the flux of carbon into this pathway, thus influencing 'sink' tissues' strength (WOODROW & BERRY, 1988; SLATTERY, KAVAKLI & OKITA, 2000; TETLOW, 2006). This approach was successfully achieved in different plant species. In barley,

AGPase was found to be allosterically unregulated, thus it would also be possible for such genes to be used for the production of high-starch yielding cultivars in other crops through genetic modification (THORBJØRNSEN *et al.*, 1996a, 1996b; RUDI, DOAN & OLSEN, 1997; DOAN, RUDI & OLSEN, 1999). The greater capacity of plants to store fixed carbon can be successfully targeted as a breeding objective by increasing 'sink' strength, which was demonstrated in maize (GIROUX *et al.*, 1996) and potato (STARK *et al.*, 1992) using forms of AGPase that were allosterically unregulated. Heat sensitivity stabilisation of AGPase was also demonstrated in maize, which resulted in improved yield stability under heat stress (GREENE & HANNAH, 1998a). In wheat and rice, transgenic up-regulation of AGPase activity through expression of maize AGPase leads to an increased total biomass and more than 20% increase in seed yield, mainly because of the increased number of seeds per plant (SMIDANSKY *et al.*, 2002, 2003). Similar results of increased starch production were obtained in rice by the expression of an allosterically insensitive highly active (up to 13-fold higher) *Escherichia coli glgC* triple mutant AGPase gene in the cytosol, which resulted in an up to 11% increase in seed weight (SAKULSINGHAROJ *et al.*, 2004).

Up-regulation of AGPase activity in wheat and rice by transformation with a modified *Sh2* maize AGPase large subunit sequence (*Sh2r6hs*) led to an increased seed weight per plant and total plant biomass increase of respectively 31% and 38% in wheat, and of 23% and 22% in rice. Harvest index, average individual seed weight and their protein or starch contents were largely not affected, and the higher yield came primarily because of increased seed number per plant. It was explained by increased allocation of resources to developing seeds and thus leading to greater numbers of seeds per head that avoided abortion (SMIDANSKY *et al.*, 2002, 2003; MEYER *et al.*, 2004). A similar result was achieved through transformation with unregulated AGPase in potato that allowed a higher starch content (STARK *et al.*, 1992). The higher level of starch synthesis may reciprocally stimulate more efficient photosynthesis by pulling greater amounts of sugars into seeds, reducing inhibition

power of sugars on photosynthesis in leaves (CHOI *et al.*, 1998; SUN, OKITA & EDWARDS, 1999).

Apart from AGPase regulation, sink strength was successfully enhanced by overexpression of apoplastic invertase (SONNEWALD *et al.*, 1997; WEBER *et al.*, 1998) under the control of a meristem-specific promoter in *Arabidopsis*, which resulted in a more than 20% increase in seed yield (HEYER *et al.*, 2004). Sucrose transporters are another promising target for sink strength increase because they are involved with assimilate unloading (CAMP, 2005). Their overexpression in potato plants leads to a higher concentration of sugars in tubers (LEGGEWIE *et al.*, 2003). Threefold to fourfold higher levels of sugars that resulted from 16-fold higher export rates of sucrose from leaves were achieved in tobacco. It led to improved growth, increased seed yield and pathogen resistance. It was done through overexpression of the maize-derived pathogenesis-related (PR-1) protein in tobacco plasmodesmata, which resulted in increased symplastic sucrose transport (STITT, 1996; MURILLO *et al.*, 2003). In other biotechnological studies with potato, a 39% increase in tuber yield and a 60% increase in its starch content was a result of the downregulation of the adenylate kinase plastidial isoform (REGIERER *et al.*, 2002). Phytochrome overexpression with its positive effect on photosynthesis and tuber yield was demonstrated in potato (BOCCALANDRO *et al.*, 2003). In rice, an overexpression of SYT1 and STZ genes enhances seed size and seed yield (SAKAMOTO & MATSUOKA, 2004; CAMP, 2005).

As can be seen, conventional plant breeding and biotechnological methods can be successfully used to alter and improve both the quality and the quantity of starch in cereals, which are to be used as a feedstock for ethanol production (BURRELL, 2003). Increase in the quantity of stored starch and sugars in storage organs (e.g. seeds) altered by breeding can be expected to directly affect fermentable substance yield per unit of area. However, yield improvements achieved at individual plant level would not necessarily transpose to better plant performance in a competitive, resource-limited environment under field conditions calculated per unit area, therefore large-

scale field trials will be required to evaluate the achieved effect in industrial conditions (MEYER *et al.*, 2004; CAMP, 2005).

2.10. CONCLUSIONS

Triticale creation and improvement has a history of nearly 135 years. Many research centres around the globe contributed to the development of this new crop. An important step in hexaploid triticale breeding is the hybridisation of octoploid and hexaploid triticale and the creation of a principally new so-called secondary hexaploid triticale. The most useful crossing schemes in triticale breeding are [bread wheat / rye // hexaploid triticale], and [triticale 'A' / bread wheat // triticale 'B']. With the expansion of triticale variability comes the increased importance of inter-line crosses [triticale / triticale]. The last 20–30 years of triticale breeding are marked with considerable progress, which is linked to effective recombinant selection based on accumulated genetic polymorphism.

The average cereal grain is low in protein and high in carbohydrate content, which is important for its use as a raw stock for ethanol production. Starch content in the grain varies depending on the genotype and environmental conditions in a wide range. Soil water deficits decrease the content of both starch and amylose in grain, but increase protein content. Amylose content has a substantial genetic variability and naturally ranges from 0% to about 40–50% in major cereals. Starch structures are also influenced by growth temperature, which may change the amylose/amylopectin ratio, the molecular structure of amylase and amylopectin, and the distribution of the amylopectin chain length. Higher temperatures during cereal grain filling result in reduced starch synthesis (lesser starch content in endosperm, smaller starch granules), a higher amylose content and a higher temperature of starch gelatinisation. In addition to starch, cereal endosperm contains considerable amounts of non-starch polysaccharides (*ca*. 10–20%), which are deposited in the endosperm cell walls. The starch content of grain is negatively correlated to protein and the non-starch

polysaccharide content. Ethanol output and yield from triticale grain is often higher than that from rye or wheat.

Starch is not deposited in plant cells in a homogenous form, but as semi-crystalline starch granules. The internal amorphic phase of starch granule is composed of amylose. High-amylopectin (waxy) starches are more easily digestible than normal or high-amylose starches. A-type crystallite polymorphic starch is easily digestible, in contrast to the B-type and some C-type starches, which are very resistant to enzymatic hydrolysis. Disc or lenticular-shaped A-type starch granules of triticale are easily hydrolysed by amylases because they have loosely packed internal structures, in contrast to smaller B-type granules. The B-type starch granules show lower (usually 2–3% less) amylose content and a higher level of phospholipids and granule-associated proteins compared to A-type granules. Higher levels of nitrogen fertilisation lead to an increased percentage of B-type granules in starch. Large A-type starch granules are more predominant in genotypes with soft than those with hard kernels. Smaller B-type starch granules have a tendency to be more susceptible to environmental stresses. The percent volume of small starch granules is positively correlated with grain protein content, whereas starch and amylose content in grain is positively correlated with large starch granules' percent volume.

Starch synthesis is a complex multi-stage process, with 14 enzymes and a number of their isoforms involved. So far, over 20 known genes are involved in starch production, with minor genes (QTL) and modifiers also involved. The biochemical effects of recessive alleles of all starch-modifying mutations always (to larger or lesser extent) cause a decrease of total starch content in grain. AGPase is a main regulator of starch synthesis in plants; it controls the rate-limiting step in starch biosynthesis. Mutations of genes that control large and small AGPase subunits trigger a strong reduction in starch content. Amylose synthesis from ADP-glucose is catalysed only by starch-synthases, but amylopectin synthesis aside from being catalysed by starch-synthases is also catalysed by starch-branching and starch-debranching enzymes. Because of starch-synthases' temperature sensitivity,

endosperm filling slows down at temperatures above 20°C. Attempts to produce novel amylopectin types by the knockdown of some enzymes in the synthesis pathway did not lead to successful results. GBSS-I enzyme is critical for amylose biosynthesis – loss of GBSS-I leads to the synthesis of amylose-free (waxy) starch. A lower starch content and yield was reported for waxy wheat lines, which hinders their usage for ethanol production. Genotypes with amylose content of more than 60% were produced in wheat and potato by the simultaneous inhibition of SBEI and SBEII enzymes. Reduction of granule-associated 14-3-3 protein accumulation results in increased starch accumulation. Hereditary fixed redistribution of starch's fractional composition seems to be possible if its regulating genetic factors and genetic diversity in loci of interest are identified and followed by targeted breeding and selection. Such plant material with altered starch properties can be exploited by plant breeding to satisfy requirements for starch quality from different industries, including bio-ethanol industry.

Starch technological properties are characterised by numerous combinations of independent traits, but leading among them are the ability of starch granules to swell, the stability of starch molecular structure in the dispersed phase, the gelling ability of the starch and its suitability for digestion by amylolytic enzymes. Starch gelatinisation results in the loss of its crystalline structure resulting in increased susceptibility for amylolytic degradation. Smaller starch granules require a higher temperature for gelatinisation. High-amylopectin starches are characterised by gelatinisation at a low temperature, fast hydration, high degree of water-binding and increased susceptibility for digestion by amylolytic enzymes. On the contrary, high amylose content was observed to have a significantly adverse effect on fermentation efficiency, particularly when amylose content is above 30%. Amylose acts as an inhibitor of swelling, especially in the presence of lipids. Beside general efforts of breeding for increased starch yield, the development of high-amylopectin and high-amylose genotypes can be viewed as independent directions of starchy crop improvement.

A selection of genotypes with increased starch content requires simple, yet repeatable methods of desirable genotype identification. Starch is known to be notoriously difficult to measure, with different analytical methods giving substantially different results. The main methods employed for starch analysis in commercial practice are based on starch hydrolysis (acidic or enzymatic) followed by the quantification of the produced glucose via its measurement by polarimetry or colorimetry. As an indirect method of grain quality determination for ethanol production, protein measurement could be employed, because protein and starch contents in grain are in strong negative correlation to each other. Amylose and amylopectin are clearly distinguished by their characteristic iodine-starch reaction. Comparable results in measured amylose content were reported when size exclusion chromatography, differential scanning calorimetry, iodine-binding and lectin-binding methods were collated. The rapid screening method of plant breeding material was developed based on the iodine staining of pollen and grain, which allows for the determination of waxy genotypes and phenotypes. Near infra-red reflectance/transmittance spectroscopy (NIRS/NITS) methods can rapidly provide both physical and chemical information about a given sample and simultaneously measure a number of parameters. The NIRS/NITS methods are an alternative to conventional chemical laboratory methods. Analysis by NIRS/NITS is dependent on reliable calibration against a suitable standard method.

Ethanol output and yield can be measured using a small-scale laboratory process that emulates commercial potable ethanol production. The average empirical conversion rate of starch into ethanol by fermentation for cereals is ~90–95%. Ethanol output from the source material depends on the amount of starch and other fermentable substances in the feedstock, the conversion ratio of this starch into fermentable sugars, and the fermentation efficiency of these sugars into ethanol.

Variation in ethanol yield between sites and years is commonly larger than between cultivars. Small grain cereals with the following traits were shown to be valuable for ethanol production: highest yielding cultivars (high starch production per

hectare) with large well-filled grain, low length to width ratio, low or medium protein content, high starch content, high starch turbidity, low residue viscosity, and no fungal contamination (very low mycotoxin content). The high ethanol yield of lines with low residue viscosities can be explained by the negative correlation between non-starch polysaccharides and starch content in whole grain. Hard, vitreous grain is not preferred for distilling because of its higher protein content, lesser accessibility of starch for enzymes and higher energy requirement for milling. High-amylose and high-amylopectin cultivars are shown to be less productive than cultivars with normal starch. Low protein content is viewed as the best predictor for high ethanol output, and it is shown that starch content alone could not be used as an accurate predictor for ethanol output. Among different protein fractions, low gliadins specifically enhance ethanol output, as shown in wheat.

The biological value of triticale cultivars is comparable to the most suitable wheat cultivars for ethanol processing. In some research, triticale was shown to be the most efficient crop (compared to other small grain cereals) in terms of cost per litre of ethanol generated. An ideal crop for bio-fuel production should have a sustained capacity to capture and convert the available solar energy into harvestable biomass with maximal efficiency and with minimal inputs and environmental impacts. Because different genetic mechanisms influence ethanol yield, it could be possible to develop improved breeding lines by combining desired ethanol yield traits from complementary parents. There is a need to breed cereals with low nitrogen requirements for use in ethanol production. Gliadins are the most responsive to the level of nitrogen supply and thus have significant potential for direct selection for improved nitrogen efficiency and improved ethanol yield. A number of conventional and transgenic approaches were demonstrated to be feasible for the production of cereals with altered starch quality and its resultant higher yield. These methods could also be used in triticale breeding with the aim of improving its starch properties that would then lead to higher ethanol yields.

Chapter 3: Materials and methods

3.1. PLANT MATERIAL AND TRIAL LOCATIONS

A flowchart of the study is depicted in Figure 3.1.1. The depicted cycles of field trials, laboratory and statistical data analysis were done in the 2006–2007 and 2007–2008 seasons. The spring triticale (×*Triticosecale* Wittmack ex A. Camus) plant material used in the laboratory analysis was obtained from field trials conducted by the Stellenbosch University triticale breeding programme[*]. In the 2006 season elite breeding block trials were planted in 6 locations, namely Vredenburg, Langgewens, Mariendahl, Roodebloem, Tygerhoek and Napier (Figure 3.1.2; Table 3.1.1). In the 2007 season elite breeding block trials were expanded to 9 locations, namely Piketberg, Klipheuwel, Langgewens, Mariendahl, Roodebloem, Tygerhoek, Napier, Riversdale and Albertinia (Figure 3.1.3; Table 3.1.1). Each elite trial consisted of 20 elite entries in 4 repetitions (reduced to 3 repetitions in the 2007 season) established in a randomised complete block design (RCBD). Codified lists of elite lines of the 2006 and 2007 seasons with their pedigrees are presented in Tables 3.1.2 and 3.1.3. In addition 80 entries (60 lines plus 20 checks) of the 2006 season senior breeding block from the Mariendahl were used in laboratory analysis of moisture and starch contents for the NIRS calibration development (Table 3.1.4). No senior trials material from the 2007 season was used in this study.

[*] Plant Breeding Laboratory, Department of Genetics, Stellenbosch University, Private Bag X1, Matieland 7602, South Africa.

Figure 3.1.1. A flowchart of the study

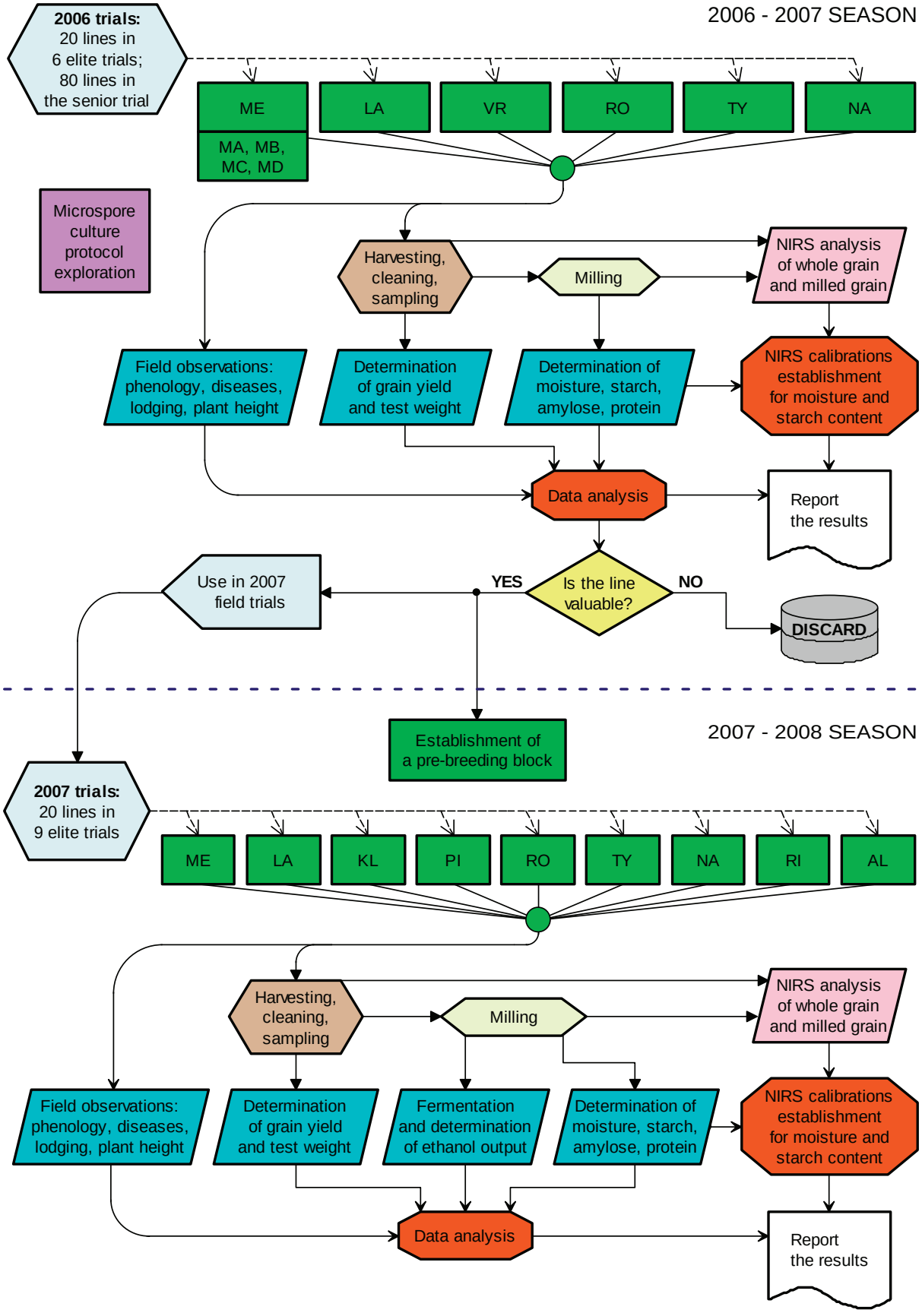
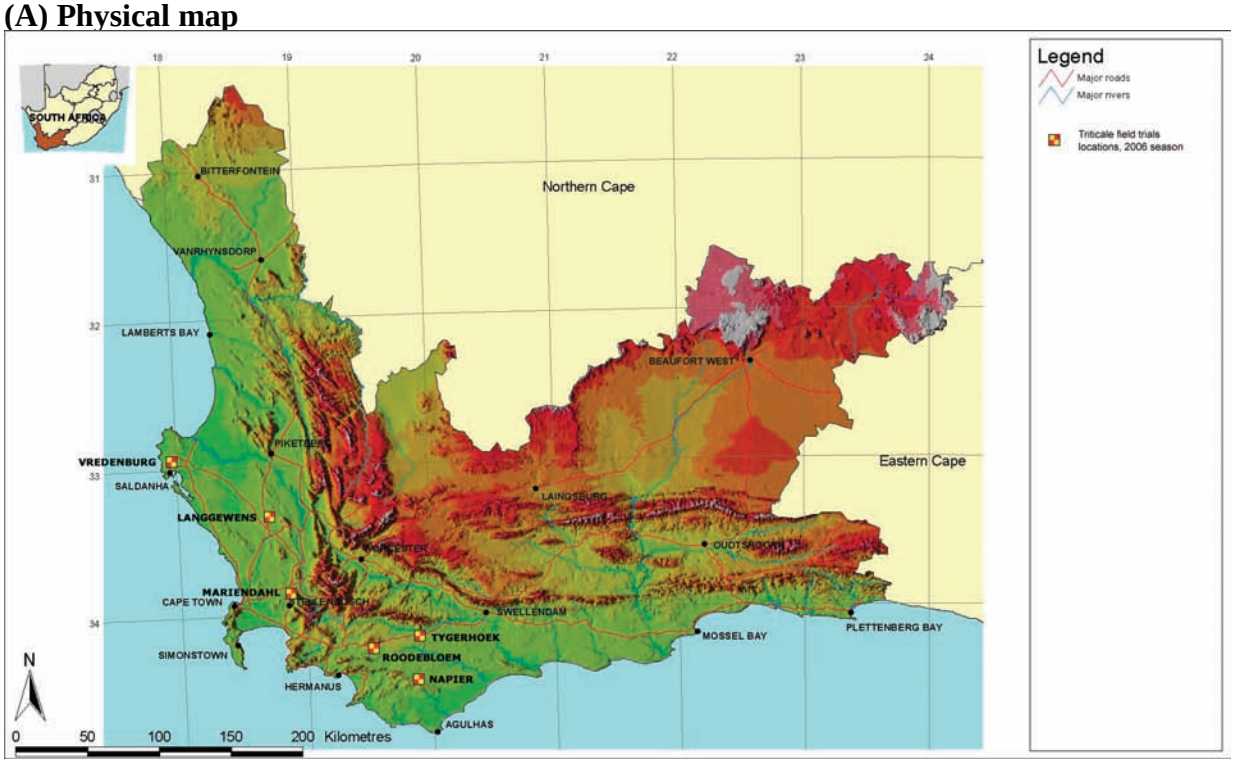


Figure 3.1.2. Locations representing triticale field trials of the 2006 season in the Western Cape cereal production area
(maps source: SA Department of Environmental Affairs and Tourism, 2007)

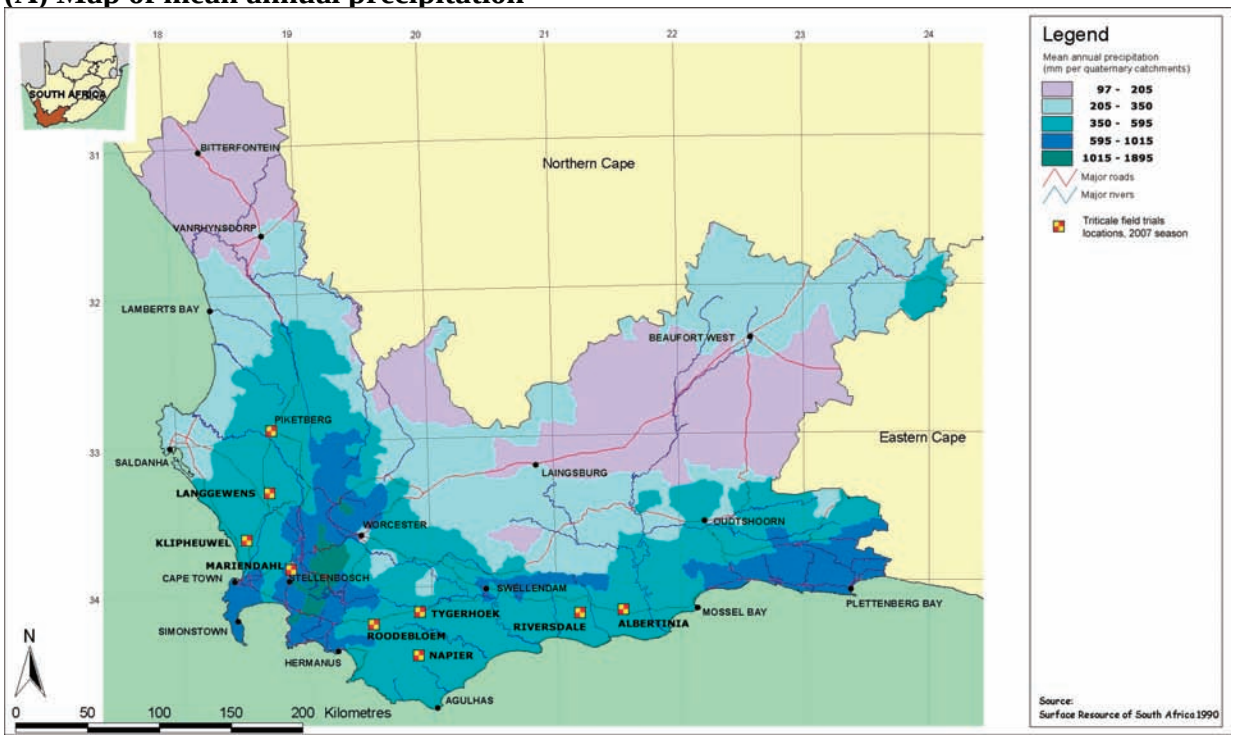
(A) Physical map



(B) Land use map

Figure 3.1.3. Locations representing triticale field trials of the 2007 season in the Western Cape cereal production area
(maps source: SA Department of Environmental Affairs and Tourism, 2007)

(A) Map of mean annual precipitation



(B) Soil types map

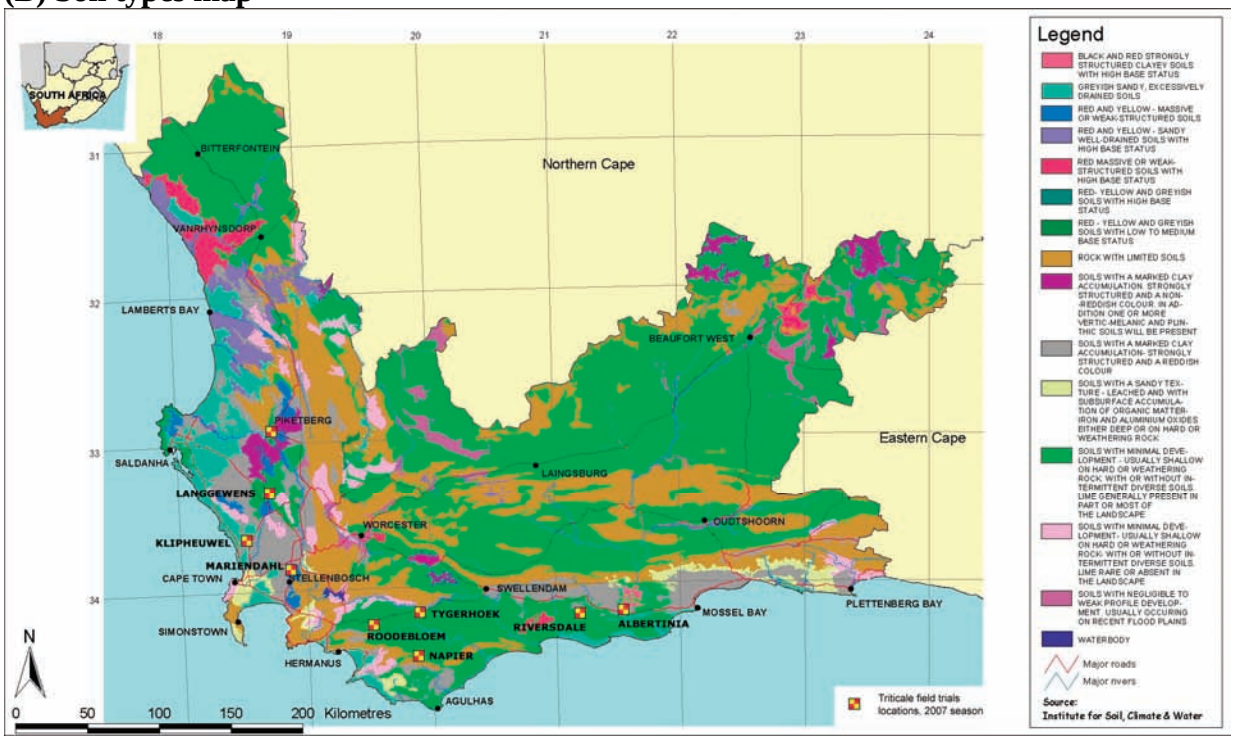


Table 3.1.1. Triticale field trials of 2006 and 2007 seasons: locations coordinates and dates of planting and harvesting

Location name	Code name	Nearest town	Farm name	Altitude above sea level, m	GPS coordinates	2006		2007	
						Planting	Harvesting	Planting	Harvesting
Swartland region									
Mariendahl	**ME** (elite); **MA, MB, MC, MD** (senior)	Elsenburg / Stellenbosch	Mariendahl Experimental Station	256	S33 82.941 E18 86.832	02-May	30-Nov	07-May	01-Dec
Langgewens	**LA**	Moorreesburg / Malmesbury	Langgewens	177	S33 16.837 E18 42.575	15-May	14-Nov	17-May	15-Nov
Vredenburg	**VR**	Vredenburg	Holvlei	45	S32 56.458 E17 56.066	11-May	08-Nov		
Klipheuwel	**KL**	Klipheuwel	Altona	162	S33 41.910 E18 42.060			15-May	13-Nov
Piketberg	**PI**	Piketberg	Panorama	171	S32 48.954 E18 50.984			07-May	15-Nov
Overberg (Ruens) region									
Roodebloem	**RO**	Caledon	Roodebloem	128	S34 14.305 E19 25.778	09-May	20-Nov	02-May	08-Nov
Tygerhoek	**TY**	Riviersonderend	Tygerhoek	183	S34 08.975 E19 54.871	04-May	22-Nov	02-May	08-Nov
Napier	**NA**	Napier	Napier Experimental Station	106	S34 28.311 E19 54.319	17-May	07-Nov	04-May	05-Nov
Riversdale	**RI**	Riversdale	Uitkyk	150	S34 05.715 E21 15.283			02-May	05-Nov
Albertinia	**AL**	Albertinia	Driefontein	200	S34 12.289 E21 35.113			02-May	05-Nov

Table 3.1.2. Pedigrees of 2006 triticale elite block entries

No.	Name*	Pedigree
1	**CA**	SUPI 3//HARE 7265/YOGUI 1
2	**CB**	ANOAS“S”
3	**CC**	TARASCA 87-1/YOGUI 1
4	**CD**	W.TCL 83/HOHI//RHINO 4/3/ARDI 1/4/KIEWIET
5	**CE**	FLORIDA 201/3/DURUM WHEAT/BALBO//BOK“S”
6	**D1**	IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
7	**DB**	DF“S”/SPD“S”(PFT80380)/4/CIT“S”/SPY/3/IA/TK//CMH73A.785/5/LYNX“S”/6/BREAD WHEAT/RHINO“S”
8	**YA**	DAHBI/5/6TA876/6TB164//PND-T/RHM/3/TESMO_2/4/2*ERIZO_12
9	**DC**	KISSA_&-3//SIKA 26/HARE_337/3/LT1478.82/FARAS_1//NIMIR
10	**YB**	RONDO/BANT_5//ANOAS_2/3/RHINO_3/BULL_1-1
11	**YC**	PRESTO//2*TESMO_1/MUSX 603/4/ARDI_1/YOPO 1419//ERIZO_9/3/SUSI_2
12	**DD**	PASSIE_3-2//GNU*2SPB/6/TEJON/BGL“S”/5/BGL DERIV SEL BULK/3/MTZ TCL/TRIGO GOOD SEED//BGL GOOD SEED/4/NUTRIA
13	**DE**	IBIS/BACCHUS
14	**DF**	FD-693/2*FAHAD_4//IBIS
15	**DG**	ANOAS 1-1/4/CHIVA”S”//YAV 79/TH JUNCEUM/3/TJ/BGL”S”/5/LASKO/2*ERIZO_11 //PANG/3/VICUNA_4
16	**DH**	RHINO 1R.1D#2/2*POLLMER_2/5/ASAD*2/JUN//ANOAS_5/3/SONNI_6/4/ASAD/ELK 54//ERIZO_10
17	**Y1**	POLLMER_2.2.1*2//FARAS/CMH84.4414
18	**Y2**	POLLMER_2.2.1*2//FARAS/CMH84.4414
19	**YD**	ERIZO_10/2*BULL_1-1//CAAL/4/2*PACA_2/COPI_1-1/3/ARDI_1/TOPO 1419//ERIZO_9
20	**Y3**	POLLMER_2.2.1*2//FARAS/CMH84.4414

* Codified due to confidentiality.

Table 3.1.3. Pedigrees of 2007 triticale elite block entries

No.	Name*	Pedigree
1	**CA**	SUPI 3//HARE 7265/YOGUI 1
2	**CC**	TARASCA 87-1/YOGUI 1
3	**CE**	FLORIDA 201/3/DURUM WHEAT/BALBO//BOK“S”
4	**CD**	W.TCL 83/HOHI//RHINO 4/3/ARDI 1/4/KIEWIET
5	**D1**	IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
6	**D2**	IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
7	**D3**	IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
8	**D4**	IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
9	**YC**	PRESTO//2*TESMO_1/MUSX 603/4/ARDI_1/YOPO 1419//ERIZO_9/3/SUSI_2
10	**Y2**	POLLMER_2.2.1*2//FARAS/CMH84.4414
11	**EA**	ANOAS 1-1/4/CHIVA“S”//YAVAROS 79/TH JUNCEUM/3/TJ/BGL“S”/5/ANOAS 1-1/REX
12	**EB**	FAHAD 5/3/LT616.82//TESMO 1/MUSX 603/4/IBIS/PAPION
13	**G1**	IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”/8/TOBIE
14	**G2**	IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”/8/TOBIE
15	**H1**	MASSA/NIMIR 3/3/YOGUI 1/TARASCA 87 3//HARE 212/4/IBIS/8/IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
16	**H2**	MASSA/NIMIR 3/3/YOGUI 1/TARASCA 87 3//HARE 212/4/IBIS/8/IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
17	**H3**	MASSA/NIMIR 3/3/YOGUI 1/TARASCA 87 3//HARE 212/4/IBIS/8/IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”

(continued)

Table 3.1.3. (continued)

No.	Name*	Pedigree
18	**H4**	MASSA/NIMIR 3/3/YOGUI 1/TARASCA 87 3//HARE 212/4/IBIS/8/IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
19	**H5**	MASSA/NIMIR 3/3/YOGUI 1/TARASCA 87 3//HARE 212/4/IBIS/8/IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
20	**H6**	MASSA/NIMIR 3/3/YOGUI 1/TARASCA 87 3//HARE 212/4/IBIS/8/IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”

* Codified due to confidentiality.

Table 3.1.4. Pedigrees of 2006 triticale senior block entries

No.	Name*	Pedigree
Block MA		
1	**CA**	SUPI 3//HARE 7265/YOGUI 1
2	**CB**	ANOAS“S”
3	**CC**	TARASCA 87-1/YOGUI 1
4	**CD**	W.TCL 83/HOHI//RHINO 4/3/ARDI 1/4/KIEWIET
5	**CE**	FLORIDA 201/3/DURUM WHEAT/BALBO//BOK“S”
6	**BA**	31ITSN180
7	**BB**	RAHUM/16ITYN7//CF6 79T70-6-2-PL1-2
8	**BC**	n/a
9	**BD**	n/a
10	**BE**	29ITSN116/30ITSN34
11	**BF**	30ITYN36/29ITSN15
12	**BG**	34ITYN8
13	**BH**	n/a
14	**BI**	23ITYN17-3/26ITYN12
15	**BJ**	23ITYN17-3/26ITYN12
16	**BK**	37ITYN7
17	**BL**	38ITYN11
18	**BM**	39ITYN25
19	**BN**	40ITYN32
20	**BO**	41ITYN39

(continued)

Table 3.1.4. (continued)

No.	Name*	Pedigree
Block MB		
1	**CA**	SUPI 3//HARE 7265/YOGUI 1
2	**CB**	ANOAS“S”
3	**CC**	TARASCA 87-1/YOGUI 1
4	**CD**	W.TCL 83/HOHI//RHINO 4/3/ARDI 1/4/KIEWIET
5	**CE**	FLORIDA 201/3/DURUM WHEAT/BALBO//BOK“S”
6	**BP**	31ITYN33//22ITYN9/REX
7	**BQ**	29ITSN69-2//24ITSN173/KIEWIET
8	**BR**	29ITSN69-2//24ITSN173/KIEWIET
9	**BS**	29ITSN69-2//28ITSN21/CF6 88T1038-3-1
10	**BT**	30ITSN113/5/W.TCL 83/HOHI//RHINO 4/3/ARDI 1/4/KIEWIET
11	**BU**	30ITSN113//CF6 88T1038-3/19ITSN70-4
12	**BV**	30ITSN196/28ITYN48
13	**BW**	22ITSN32/5/15ITYN38/GY30nr.17(VPM)//USGEN18/3/19ITYN10/4/18 ITSN95//19ITYN10/17ITSN80/6/26ITYN28/REX
14	**BX**	21ITYN14/4/CHIVA‘S’//YAR79/TH.JUNCEUM/3/17ITYN3/5/97M901 -14 10(44)-1
15	**BY**	21ITYN14/4/CHIVA‘S’//YAR79/TH.JUNCEUM/3/17ITYN3/5/97M901 -14 10(44)-1
16	**BZ**	21ITYN14/4/CHIVA‘S’//YAR79/TH.JUNCEUM/3/17ITYN3/5/16ITYN 4/18ITSN95//454.19
17	**UA**	21ITYN14/4/CHIVA‘S’//YAR79/TH.JUNCEUM/3/17ITYN3/5/16ITYN 4/18ITSN95//454.19
18	**EA**	22ITYN9/4/CHIVA‘S’//YAR79/TH.JUNCEUM/3/17ITYN3/5/28ITYN1 6//CF6 88T1128-2-2-2/REX
19	**UB**	22ITYN9/4/CHIVA‘S’//YAR79/TH.JUNCEUM/3/17ITYN3/6/22ITSN3 2/5/15ITYN38/GY30nr.17(VPM)//USGEN18/3/19ITYN10/4/18ITSN95/ /19ITYN10/17ITSN80
20	**UC**	SCR 13//21ITYN14/USGEN19-4/3/22ITYN9/CF6 88T1030-2

(continued)

Table 3.1.4. (continued)

No.	Name*	Pedigree
Block MC		
1	**CA**	SUPI 3//HARE 7265/YOGUI 1
2	**CB**	ANOAS“S”
3	**CC**	TARASCA 87-1/YOGUI 1
4	**CD**	W.TCL 83/HOHI//RHINO 4/3/ARDI 1/4/KIEWIET
5	**CE**	FLORIDA 201/3/DURUM WHEAT/BALBO//BOK“S”
6	**UD**	A3130(WINTER TYPE FROM CANADA)/97T187-A+97T131//26ITSN140/A3078(WINTER TYPE FROM CANADA)
7	**UE**	A3130(WINTER TYPE FROM CANADA)/97T187-A+97T131//26ITSN140/A3078(WINTER TYPE FROM CANADA)
8	**UF**	35ITYN24
9	**UG**	35ITSN39
10	**UH**	18ITSN33/114N92(3GC87)//28ITSN105
11	**UI**	18ITSN33/114N92(3GC87)//28ITSN105
12	**UJ**	29ITYN18/28ITYN48
13	**UK**	22ITSN41/16ITSN46-1-1//ROMANIA YIELD
14	**UM**	29ITSN131/29ITSN43
15	**UN**	29ITSN131/29ITSN43
16	**UO**	36ITSN25
17	**UP**	36ITSN57
18	**UQ**	36ITSN59
19	**UR**	36ITSN69
20	**US**	36ITSN82

(continued)

Table 3.1.4. (continued)

No.	Name*	Pedigree
Block MD		
1	**CA**	SUPI 3//HARE 7265/YOGUI 1
2	**CB**	ANOAS“S”
3	**CC**	TARASCA 87-1/YOGUI 1
4	**CD**	W.TCL 83/HOHI//RHINO 4/3/ARDI 1/4/KIEWIET
5	**CE**	FLORIDA 201/3/DURUM WHEAT/BALBO//BOK“S”
6	**UT**	36ITSN85
7	**UU**	36ITSN92
8	**UV**	36ITSN93
9	**UW**	36ITSN133
10	**UX**	36ITSN134
11	**UY**	36ITSN136
12	**UZ**	36ITSN137
13	**ZA**	36ITSN140
14	**ZB**	36ITSN142
15	**ZC**	36ITSN144
16	**ZD**	36ITSN156
17	**ZE**	22ITYN9/4/CHIVA“S”//YAR79/TH.JUNCEUM/3/17ITYN3/5/22ITSN41/16ITSN46-1-1//ROMANIA YIELD
18	**ZF**	22ITYN9/4/CHIVA“S”//YAR79/TH.JUNCEUM/3/17ITYN3/5/22ITSN41/16ITSN46-1-1//ROMANIA YIELD
19	**ZG**	30ITYN36/29ITYN43
20	**ZH**	30ITYN36/29ITYN43

* Codified due to confidentiality.

3.2. TRIALS PLANTING AND HUSBANDRY

Trials were planted under rainfed conditions during May 2006 and 2007 (Table 3.1.1). Field trials were established predominantly on canola (rapeseed, *Brassica napus* L.) as a forecrop and some on fallow land. Each plot of 5.1m^2 consisted of 6 rows spaced 17cm apart; the row length was 5m. Nitrogen fertiliser in the form of carbamide (urea) with 46% N content was applied before planting or while preparing the seedbed (with a disc plough) at a rate of 45kg.ha^{-1} N, and trials also received a top dressing of 45kg.ha^{-1} N six weeks after planting. A self-propelled Øyjord OSD plot drill (F. Walter - H. Wintersteiger K.G., Austria) was used for planting the seed at 300seeds.m^{-2}. The following herbicides were sprayed straight after planting: in the 2006 season a mixture of Hussar® (0.372kg.ha^{-1}; Bayer CropScience Pty. Ltd.), Buctril® (0.496L.ha^{-1}; Bayer CropScience Pty. Ltd.) and MCPA (1.48L.ha^{-1}; Bayer CropScience Pty. Ltd.); in the 2007 season a mixture of Hussar® (0.08kg.ha^{-1}; Bayer CropScience Pty. Ltd.), Buctril® (0.15L.ha^{-1}; Bayer CropScience Pty. Ltd.), MCPA (0.2L.ha^{-1}; Bayer CropScience Pty. Ltd.) and Ballista® (adjuvant, 0.2L.ha^{-1}; Bayer CropScience Pty. Ltd.). No insecticides or fungicides were applied.

3.3. ENVIRONMENTAL CONDITIONS

Environmental condition raw data (precipitation, maximum and minimum temperatures averaged over ten-day periods (TDP)), as well as respective long-term (LT) data for each season and location was obtained from the closest weather station (courtesy RITHA WENTZEL, Agro-Climatology, ARC Institute for Soil, Climate and Water, Stellenbosch, RSA, 2009). For the characterisation of meteorological conditions during the growth season Selyaninov's hydrothermal coefficient, HTC (SELYANINOV, 1958; KLESCHENKO, ZOIDZE & BOKEN, 2005; POTOP & SOUKUP, 2009) was calculated at the end of every TDP as a running average with a frame period of three TDP:

$$HTC_k = \frac{\left(\frac{\sum P_{k-2}}{0.1\sum_{T\geq 10} T_{k-2}} + \frac{\sum P_{k-1}}{0.1\sum_{T\geq 10} T_{k-1}} + \frac{\sum P_k}{0.1\sum_{T\geq 10} T_k} \right)}{3}$$

where $\sum P_k$, $\sum P_{k-1}$, $\sum P_{k-2}$ – cumulative precipitation (mm) of the given TDP *k*, previous TDP *k-1* and the TDP prior to the previous TDP *k-2*, respectively; $\sum_{T\geq 10} T_k$, $\sum_{T\geq 10} T_{k-1}$, $\sum_{T\geq 10} T_{k-2}$ – the sum of the average daily air temperatures (°C) for the same respective periods *k*, *k-1* and *k-2* with daily mean temperatures above 10°C. Values of HTC were interpreted according to Table 3.3.1.

Table 3.3.1. Selyaninov's hydrothermal coefficient (HTC) values interpretation

Colour code	HTC value	Description
	0.19 or less	extremely severe drought
	0.20 – 0.39	severe drought
	0.40 – 0.60	moderate drought
	0.61 – 0.75	weak/mild drought
	0.76 – 1.00	dry, incipient dry spell
	1.01 – 1.30	insufficiently wet, near normal
	1.31 – 1.60	slightly wet/moderately wet
	1.61 – 1.90	very wet
	1.91 or more	extremely wet

3.4. FIELD OBSERVATIONS AND HARVESTING

During both seasons trials were visited on a regular basis in order to make *in situ* observations and record agronomic data for plant height (cm), phenological stage (days from planting to heading), scoring of lodging, and adult plant resistance to leaf rust (*Puccinia triticina* Eriks.) and stem rust (*Puccinia graminis* Pers.:Pers.). Disease resistance was recorded according to Table 3.4.1 (CIMMYT, 2006). The field trials were harvested with a Wintersteiger Nurserymaster Elite (Wintersteiger Gesellschaft m.b.H. & CO, Ried, Austria) and a Hege 125C (Hans Ulrich Hege Maschinenbau, Hohebuch, Western Germany) field plot harvester during November 2006 and 2007 (Table 3.1.1). Each plot was harvested *in toto*. The seeds were cleaned with a seed cleaner Hub-O-Mat SC 800-12 (K. Huber Engineering, RSA)

and kept for subsequent analysis. Laboratory analysis of grain yield ($kg.ha^{-1}$) and test weight ($kg.HL^{-1}$; measured using a 0.5L hectolitre mass meter (A.F.H. Devers & Co.(Pty.)Ltd., Johannesburg, RSA) based on AACC Method 55-10) from each entry was performed and data recorded.

Table 3.4.1. Major severity and field response classes for stem rust and leaf rust (CIMMYT, 2006)

Infection type*	Host response; symptoms
0	No visible infection on plants.
R	**Resistant**; visible chlorosis or necrosis, *no uredia* are present.
MR	**Moderately Resistant**; *small uredia* are present and surrounded by either chlorotic or necrotic areas.
M	**Intermediate**; *variable sized uredia* are present, some with chlorosis, necrosis, or both.
MS	**Moderately Susceptible**; *medium sized uredia* are present and possibly surrounded by chlorotic areas.
S	**Susceptible**; *large uredia* are present, generally with little or no chlorosis and no necrosis.

* Detailed outlines for recording stripe, stem and leaf rust intensities in cereals based upon **severity** (percentage of rust infection on the plants) and **field response** (type of disease reaction) have been developed by Loegering. Severity is recorded as a percentage, according to the modified Cobb scale. This recording process relies upon visual observations and it is common to use the following intervals: **Trace, 5, 10, 20, 40, 60, 100** percent infection.

Severity and **field response** readings are usually combined. For example:

tR = Trace severity with a resistant field response.

5MR = 5% severity with a moderately resistant field response.

60S = 60% severity with a susceptible field response.

3.5. SAMPLING AND SAMPLE PREPARATION

Representative 70g samples were taken from the 2006 season trials for the subsequent determination of moisture and starch for the NIRS calibration development. A sample was

taken from a random repetition of each entry and of each location (total of 120 samples of the elite trial) and from each entry of four blocks (MA, MB, MC and MD) of the 2006 season senior trial (total of 80 samples). It resulted in a total of 200 individual triticale grain samples.

In the 2007 season, plant material was represented by 220 individual triticale grain samples (100g per entry) from the elite trials only. Due to financial constrains and time limitations not all repetitions from every trial locations were analysed. The Mariendahl elite trial material was represented by all 3 repetitions of the 20 entries (a total of 60 samples), while from the other 8 locations only 2nd repetition was used (20 samples from each location, a total of 160 samples). Whole grain samples were subjected to near infra-red spectrometer for spectral data recording.

Contents in terms of moisture, total starch, amylose-in-starch, protein (in the 2006 season), and ethanol output was determined on milled grain samples. For ethanol output determination bulked whole grain samples from all 3 repetitions of each 2007 elite trial location were taken and their moisture content standardised to 14% prior to milling. Moisture content of grain samples was determined via the conductivity method on the crop moisture detector model G-6C (Delmhorst Instrument Company, Towaco, N.J., USA) and the moisture content was adjusted to 14% by mixing it with the necessary amount of distilled water in an air-tight container 24 hours prior to milling. The grain samples were milled on the Tecator Cyclotec 1093 sample mill (Tecator, Sweden) to pass through a 0.5mm screen sieve. Each sample was kept in an individual air-tight plastic bottle (before and after milling) to prevent moisture content fluctuations.

3.6. MOISTURE, PROTEIN AND PSI DETERMINATION

Moisture content (%) of samples was determined after milling using the gravimetric method on the Denver Instrument IR35M-000230V1 moisture scale (Denver Instrument,

Germany). A 3g milled grain sample was taken and evenly spread on the moisture scale plate. The drying temperature was set at 110°C, and the drying period on "Automatic shut-down."

Protein content in whole grain (% at 12% moisture basis) and particle size index (PSI, %) were determined in the 2007 season[§], raw data courtesy of Dr MARENA MANLEY group, Department of Food Science, Stellenbosch University, RSA (DU PISANI, 2009). A NIRS method was implemented for the protein and PSI determination using Bruker MPA FT-NIR spectrophotometer (Bruker Optics GmbH, Germany), as well as BÜCHI NIRFlex N-500 Fourier transform near infra-red (FT-NIR) spectrophotometer (BÜCHI Labortechnik AG, Flawil, Switzerland).

3.7. TOTAL STARCH CONTENT DETERMINATION

Total starch content in whole grain (%, dry weight basis) was determined by an α-amylase/amyloglucosidase (AA/AMG) method (AACC Method 76.13, AOAC Method 996.11 and ICC Standard Method No. 168 according to MCCLEARY, GIBSON & MUGFORD, 1997) using the Megazyme total starch assay kit K-TSTA 05/06 (Megazyme International Ireland Ltd., Ireland). A standardised 96% (DWB) regular maize starch reference sample was included in each analysis batch. A 100mg milled grain sample was wetted with 0.2mL of 80% ethanol and treated with 3mL ($100U.mL^{-1}$) of thermostable α-amylase in a MOPS buffer (50mM, pH 7.0; 1.155% MOPS sodium salt, 0.074% (5mM) $CaCl_2.2H_2O$ and 0.02% (w/v) sodium azide in distilled water) to partially hydrolyse the starch. The tube was incubated for 6min in a boiling water bath. After completely dissolving the starch, dextrins were quantitatively hydrolysed to glucose by amyloglucosidase. The tube was placed in a bath at 50°C, and 4mL of sodium acetate buffer (200mM, pH 4.5; 1.18% glacial acetic acid ($1.05g.mL^{-1}$) and 0.02% (w/v) sodium azide in distilled water) plus 0.1mL ($3300U.mL^{-1}$) of

[§] Protein analysis for the 2006 season trials was done in an outsourced laboratory; the acquired data was not usable due to unrepeatable results.

amyloglucosidase. After the tube was incubated at 50°C for 30min, its content was diluted with distilled water to a 100mL volume and an aliquot of this solution was centrifuged at 3000RPM for 10min. A 0.1mL duplicate aliquots of the supernatant was transferred to two tubes and 3.0mL of GOPOD reagent (glucose oxidase (>12U.mL^{-1}), glucose peroxydase (>0.65U.mL^{-1}) and 4-aminoantipyrine (0.08mg.mL^{-1}) in a potassium phosphate buffer (1M, pH 7.4) plus *p*-hydroxybenzoic acid (0.22M) and sodium azide (0.02% w/w) in distilled water) were added to each tube, including reagent blank (0.1mL of distilled water) and quadruplicate glucose controls (0.1mL of D-glucose standard solution (1mg.mL^{-1}) in 0.2% (w/v) benzoic acid), and incubated at 50°C for 20min. The absorbance at 510nm was read against the reagent blank on the Spectronic 601 spectrophotometer (Milton Roy Company, USA) to determine the glucose concentration, and the Megazyme® Mega-Calc™ (Excel®-based calculator) was used for total starch calculations from the absorbance values.

3.8. AMYLOSE CONTENT DETERMINATION

Amylose-in-starch content (amylose/amylopectin ratio) was estimated by a concanavalin A (Con A) method (GIBSON, SOLAH & MCCLEARY, 1997) using the Megazyme amylose/amylopectin assay kit K-AMYL 04/06 (Megazyme International Ireland Ltd., Ireland). A starch reference sample with a specified content of amylose was included in each analysis batch. As starch pre-treatment (section 'A'), a 0.025g flour sample was wetted with 1mL of dimethyl sulphoxide (DMSO) and the tube was heated in a boiling water bath for approximately 16min, with intermittent high-speed stirring on a vortex mixer. The tube was stored at room temperature for 5min and a total of 6mL of 95% ethanol was added. The tube was left to stand for 15min to allow for starch precipitate formation, after which the tube was centrifuged at 2000g (3000RPM) for 5min. The supernatant was discarded and the tube was drained for 10min. 2mL of DMSO were added to the starch pellet and the tube was placed in a boiling water bath for 15min. 4mL of Con A solvent (30% solution of concentrated Con A

solvent (600mM, pH 6.4 sodium acetate buffer; 4.92% anhydrous sodium acetate with 17.55% NaCl, 0.05% $CaCl_2.6H_2O$, 0.07% $MgCl_2.6H_2O$ and 0.07% (w/v) $MnCl_2.4H_2O$) in distilled water) was added. The tube contents were transferred into a 25mL volumetric flask and diluted to its volume with Con A solvent, which formed Solution A. The solution was filtered through Whatman No. 1 filter paper. From this point onwards analysis was performed using two separate sets of tubes for Con A precipitation of amylopectin and determination of amylose, and determination of total starch.

For the Con A precipitation of amylopectin and determination of amylose (section 'B'), 1.0mL of Solution A was transferred to a 2.0mL Eppendorf® tube and 0.5mL of Con A solution (2mg of crystalline Con A in 0.5mL Con A solvent) was added. The tube was left to stand for 1 hour at room temperature, and then centrifuged at 14000RPM for 10min. 1mL of the supernatant was transferred to a 15mL tube, 3mL of sodium acetate buffer (100mM, pH 4.5) was added and the lightly stoppered tube was heated in a boiling water bath for 5min to denature the Con A. The tube was placed in a water bath at +40°C for 5min, after which 0.1mL of AMG/α-AMY enzyme mixture (amyloglucosidase ($330U.mL^{-1}$) plus fungal α-amylase ($50U.mL^{-1}$) in 100mM, pH 4.5 sodium acetate buffer) was added and incubated at +40°C for 30min. The tube was centrifuged at 3000RPM for 5min, and 1mL of supernatant was transferred to a glass test tube ('B') with a screw top.

For the determination of total starch (section 'C'), 0.5mL of Solution A was mixed with 4mL of sodium acetate buffer (100mM, pH 4.5) and 0.1mL of AMG/α-AMY enzyme mixture (amyloglucosidase ($330U.mL^{-1}$) plus fungal α-amylase ($50U.mL^{-1}$) in 100mM, pH 4.5 sodium acetate buffer) was added and incubated at +40°C for 10min. 1mL aliquot was transferred to a glass test tube ('C') with a screw top.

At this point (section 'D'), reagent blank (1mL of sodium acetate buffer, 100mM, pH 4.5) and duplicate glucose controls (0.1mL of D-glucose standard solution ($1mg.mL^{-1}$) with 0.9mL of 100mM, pH 4.5 sodium acetate buffer) were prepared. 4mL of GOPOD reagent

(glucose oxidase (>12U.mL^{-1}), glucose peroxydase (>0.65U.mL^{-1}) and 4-aminoantipyrine (0.08mg.mL^{-1}) in a potassium phosphate buffer (1M, pH 7.4) plus *p*-hydroxybenzoic acid (0.22M) and sodium azide (0.02% w/w) in distilled water) was added to each test tube (solutions from sections 'B' and 'C'), including reagent blank and glucose controls. Tubes were incubated concurrently at +40°C for 20min, and then the absorbance values of each sample was read at 510nm against the reagent blank on the Spectronic 601 spectrophotometer (Milton Roy Company, USA). For the calculation of amylose-in-starch content (w/w, %) the following formula was used:

$$\text{Amylose}(\%) = \frac{\text{Con A supernatant absorbance, solution 'B'}}{\text{Total starch aliquot absorbance, solution 'C'}} \times 66.8$$

3.9. ETHANOL OUTPUT, ETHANOL YIELD AND AAQ DETERMINATION

A small scale fermentation experiment using full factorial design with 3 repetitions (sampling procedure see in section 3.5 "Sampling and sample preparation") was performed for the determination of ethanol output (*EO*; L.tonne^{-1} dry matter) with (+E) and without (–E) the addition of technical saccharifying enzymes (fermentation process based on adapted protocol from SENN & PIEPER, 2001). The +E and –E data was used for the consequent calculation of the auto-amylolytic quotient (AAQ, %) and ethanol yield (L.ha^{-1} dry matter). Only samples from the 2007 elite trials were used. A 20g sample of milled triticale grain (20% w/v adjusted to 14% moisture content before milling; 17.2°Plato) was added to a 200mL serum bottle with stirring magnet. 100mL of citrate buffer (100mM, pH 3.6; a 685mL of 100mM (21.01g.L^{-1}) citric acid monohydrate ($C_6H_8O_7.H_2O$) solution mixed with a 315mL of 100mM (29.41g.L^{-1}) tri-sodium citrate dihydrate ($C_6H_5O_7Na_3.2H_2O$) solution) was added. For the E+ determination, at this point 1mL of OPTIMASH™ VR enzymes (bacterial xylanase/cellulase (1–5%) for hydrolysation of cellulose and hemicellulose in whole-grain triticale feedstock; GENENCOR, 2007) was also added. The bottle was sealed with a rubber

stopper and incubated for 2 hours at 57°C on a magnetic stirrer. Then the bottle was cooled to 30°C and 10mL of yeast (*Saccharomyces cerevisiae* L.) D5α strain was added. For the E+ determination, 1mL of STARGEN™ 002 enzymes (blend of fungal glucoamylase (10–15%) and α-amylase (1–5%) for hydrolysation of raw, uncooked granular starch; GENENCOR, 2005b; WANG *et al.*, 2007) was also added for the simultaneous saccharification and fermentation (SSF) to take place in the mash. A one-way valve on syringe was inserted into the bottle through the rubber stopper. The bottle was weighted (*W1*, g) and incubated for 72 hours at 30°C on a magnetic stirrer. At the end of the fermentation the bottle was weighted (*W2*, g) to determine the loss of CO_2 as an indirect measure of ethanol produced. A 2mL sample of the fermented solution was taken and kept frozen at –80°C for future reference. Calculations of ethanol output *EO* (–E or +E; L.tonne^{-1} as-is) were done according to the formula: $EO = (W1 - W2) \times 50 \times 1.267427$.

The AAQ was calculated as the ratio between *EO* –E and *EO* +E (%).

The ethanol yield (*EY*; L.ha^{-1} as-is) was calculated from *EO* +E (L.tonne^{-1} as-is) and the grain yield (kg.ha^{-1} as-is) as their product.

Relative ethanol output (REO, given as percentage of theoretical values calculated from total starch content) was calculated as: $REO = (EO / TEO) \times 100$, where the *TEO* is the theoretical ethanol output from starch. The *TEO* was calculated as 0.72L of ethanol with a density of 0.789kg.L^{-1} acquired from 1kg of total starch (SMITH *et al.*, 2006).

3.10. NEAR INFRA-RED REFLECTANCE SPECTROSCOPY (NIRS) DATA ACQUISITION

For NIRS analysis, a total of 200 samples from 2006 season and 220 samples from the 2007 season were used. Each sample was subjected to NIR spectroscopy analysis twice: as whole kernels and again after milling. The BÜCHI NIRLab N-200 Fourier transform near infra-red (FT-NIR) spectrometer equipped with measurement cell MCS 100 with a rotating sample desk was used (BÜCHI Labortechnik AG, Flawil, Switzerland). The samples were

presented to the spectrometer in rotating glass Petri dishes and their reflectance spectra were recorded at a fixed resolution of $8cm^{-1}$ in the near infra-red spectral range of 1000–2500nm with 0.96nm intervals (1557 data points). The NIRCal 4.21 software (BÜCHI Labortechnik AG, Flawil, Switzerland) was used for raw data storage and export to Microsoft Excel 2003 electronic tables (spectral data values converted into absorbance, Log R^{-1}). Chemometric analysis of NIR spectra together with moisture and starch data was performed for the 2006 and 2007 datasets and calibration and prediction models were developed using partial least squares regression (PLSR) models with a single response variable (PLS1), i.e. moisture content or starch content. In the pre-treatment of data, multiplicative scatter correction (MSC), Savitzky-Golay first and second derivatives (2^{nd} and 3^{rd} polynomials respectively, 11 point segment), as well as their combinations were applied. Data values of the response variables and spectra were standardised (divided by their standard deviation) and centred. The optimum PLS1 calibration models were selected based on the highest squared correlation coefficient of validation (R^2), lowest root mean square error of prediction (RMSEP) and standard error of performance (SEP), highest RPD (relative predictive determinant or the ratio of performance to deviations, RPD = standard deviation divided by SEP), and least number of PLS factors obtained after cross-validation of half of the samples by the other half. The reference data was sorted (ascending) for each compound prior to analysis, and the complete dataset split in two segments of equal size by selecting samples interleaving each other. Explicit outliers were excluded from the models considering their T-U scores from X-Y relation outliers' plots, as well as X-residual and Y-residual sample variance plots (NAES & ISAAKSSON, 1992; ESBENSEN, 2002).

3.11. STATISTICS AND DATA ANALYSIS

Statistic methods and models used for the analysis of the field and laboratory data are described in Table 3.11.1.

Table 3.11.1. Statistic methods and models used for the data analysis

Method or model; Software used	Brief description and application
Initial data entry and export: • **Microsoft Excel 2003** (Microsoft Corporation) • **KyPlot 2.0 Beta 15 32-bit** (Koichi Yoshioka, Japan) • **NIRCal 4.21** (BÜCHI Labortechnik AG, Flawil, Switzerland)	Raw data entry, processing, transformation, *ad hoc* formula calculations, dataset preparation for export to other applications.
Data plotting, graphs: • **Microsoft Excel 2003** (Microsoft Corporation) • **KyPlot 2.0 Beta 15 32-bit** (Koichi Yoshioka, Japan) • **BioStat Version 2008 Build 5.4.0.0** (AnalystSoft) • **Biplot 1.1 Excel add-in** (Eric Smith & Ilya Lipkovich, Statistics Department of Virginia Tech, USA) • **MeeSoft Diagram Designer Version 1.21.2 2008** (MeeSoft, Michael Vinther) • **Adobe Photoshop CS2 Version 9.0** (Adobe Systems Inc.)	Visualisation of numerical data in graphical form (graphs and charts) for quick grasping and exploration purposes.
Descriptive statistics: • **Microsoft Excel 2003** (Microsoft Corporation) • **SSC-Stat V2.18 Excel add-in** (Statistical Services Centre, The University of Reading, United Kingdom) • **Gnumeric 1.9.8** (Jody Goldberg & Miguel de Icaza)	General data assessment prior to analysis.
Normality tests: • **OpenStat Version 12.05.09** (William G. Miller) • **BioStat Version 2008 Build 5.4.0.0** (AnalystSoft)	Assessment of the data to determine whether it follows a normal distribution, which is required by parametric statistic methods.
ANOVA and rank indices: • **Microsoft Excel 2003** (Microsoft Corporation) • **Gnumeric 1.9.8** (Jody Goldberg & Miguel de Icaza) • **GGEbiplot Pattern Explorer 6.0** (Weikai Yan) • **The Unscrambler 9.2** (CAMO Process AS, Norway)	Analysis of the variance of factorial experiments, and multiple comparisons of samples.

(continued)

Table 3.11.1. (continued)

Method or model; Software used	Brief description and application
Spearman' rank correlation: • **KyPlot 2.0 Beta 15 32-bit** (Koichi Yoshioka, Japan)	A distribution-free (nonparametric) correlation analysis method, which does not require normally distributed data.
Cluster analyses: • **Tanagra Data Mining 1.4.31** (Ricco Rakotomalala, Lyon, France)	Multivariate nonparametric method of dimensionality reduction. Standard hierarchical agglomerative clustering strategy (Ward's criterion) using distance normalisation by variance was employed. It stratifies cases (samples, genotypes, environments etc.) into homogeneous groups, maximising the variations between them. Used to suggest genetic relationships, and to give a better understanding of environmental factors influencing adaptation. Used in a systematic approach for choosing testing sites (CROSSA & CORNELIUS, 1997).
Spatial analysis – mixed model, restricted maximum likelihood (REML): • **CropStat for Windows 7.2.2007.3 Release Build 2008.3** (Crop Research Informatics Laboratory of International Rice Research Institute, Metro Manila, Philippines)	Two-dimensional spatial analysis of the field experiments using a first-order autoregressive (AR1) correlation model of neighbouring plots in the direction of the rows and columns (AR1×AR1). It obtains balanced least squares mean fixed values which are corrected for spatial effects, thus having a reduced error level.
Additive main effects and multiplicative interaction (AMMI) model: • **CropStat for Windows 7.2.2007.3 Release Build 2008.3** (Crop Research Informatics Laboratory of International Rice Research Institute, Metro Manila, Philippines)	Cross-site (multiple locations and/or years) analysis of genotypes for genotype-by-environment (G×E) interactions. Fitted values are obtained, which have been corrected for G×E interaction effects. AMMI may provide more reliable estimates of genotype performance than the mean across sites. Enables one to identify target breeding environments and to choose representative testing sites in those environments. Helps in selection of varieties with good adaptation to target breeding environments. Can be used to identify key agro-climatic factors, disease and insect pests, and physiological traits that determine adaptation to environments (YAN & HUNT, 1998).

(continued)

Table 3.11.1. (continued)

Method or model; Software used	Brief description and application
Principal components (PC) interaction biplot: • **Biplot 1.1 Excel add-in** (Eric Smith & Ilya Lipkovich, Statistics Department of Virginia Tech, USA) • **CropStat for Windows 7.2.2007.3 Release Build 2008.3** (Crop Research Informatics Laboratory of IRRI, Metro Manila, Philippines)	Multivariate projection and dimensionality reduction technique used for graphical representation of the AMMI models. Interaction principal component axes (IPCAs) scores are extracted by singular value decomposition. Helps visualise relationships among genotypes and environments, shows both main and interaction effects. To adequately explain patterns in the data usually only the first two principal component axes (IPCA1 and IPCA2) are needed. General interpretation guidelines: genotypes that occur close to particular environments on the IPCA2 *vs*. IPCA1 biplot show specific adaptation to those environments; a genotype that falls near the center of the biplot (small IPCA1 and IPCA2 values) may have broader adaptation (ANNICCHIARICO, 2002).
Principal components analysis (PCA): • **Biplot 1.1 Excel add-in** (Eric Smith & Ilya Lipkovich, Statistics Department of Virginia Tech, USA) • **The Unscrambler 9.2** (CAMO Process AS, Norway)	Multivariate projection and dimensionality reduction, nonparametric technique for analysis of variables interrelations/correlations, e.g. investigation of genotypes and environments grouping and trends. Mainly used as an exploratory method (SHLENS, 2009). Useful for determining which agro-climatic or biotic factors can be used to discriminate among environments or genotypes. Variables were centred and standardised prior to analysis.
Partial least squares regression (PLSR): • **The Unscrambler 9.2** (CAMO Process AS, Norway)	Multivariate soft-modelling method popular in industrial applications such as chemometrics (computational chemistry), which generalises and combines features of principal components analysis and multiple linear regression. Useful for development of predictive models for response variables from a large explanatory data matrix, e.g. in spectroscopic, genomics, proteomics applications. It is able to handle data where the number of explanatory variables is much greater than the number of observations (small sample sizes); with many, noisy, multi-collinear, and missing value variables. Handles nominal, ordinal, and continuous variables. It is a nonparametric method. Because the distribution of partial least squares is unknown, there is no conventional significance test (WOLD, SJÖSTRÖM & ERIKSSON, 2001; CROSSA, CORNELIUS & YAN, 2002; ABDI, 2007).

3.12. ESTABLISHMENT OF MARS PRE-BREEDING BLOCK

Twenty three disease resistant and/or high-starch and/or high-ethanol yielding lines were selected for the establishment of marker-assisted recurrent selection (MARS) pre-breeding block (Table 3.12.1). A total of 70 heads was manually emasculated during May 2008 and crosses were performed in random order. The tillers were propagated according to the method described by MARAIS, BOTES & LOUW (2001). Created F_1 lines were planted in the 2008 junior breeding block field trials.

Table 3.12.1. List of lines used for the establishment of the marker-assisted recurrent selection pre-breeding block

No.	Name*	Pedigree
1	**CA**	SUPI 3//HARE 7265/YOGUI 1
2	**CE**	FLORIDA 201/3/DURUM WHEAT/BALBO//BOK“S”
3	**CD**	W.TCL 83/HOHI//RHINO 4/3/ARDI 1/4/KIEWIET
4	**D2**	IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
5	**D3**	IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
6	**YC**	36ITYN27
7	**Y2**	36ITSN139
8	**EA**	ANOAS 1-1/4/CHIVA“S”//YAVAROS 79/TH JUNCEUM/3/TJ/BGL“S”/5/ANOAS 1-1/REX
9	**EB**	FAHAD 5/3/LT616.82//TESMO 1/MUSX 603/4/IBIS/PAPION
10	**G1**	IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”/8/TOBIE
11	**H4**	MASSA/NIMIR 3/3/YOGUI 1/TARASCA 87 3//HARE 212/4/IBIS/8/IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
12	**H5**	MASSA/NIMIR 3/3/YOGUI 1/TARASCA 87 3//HARE 212/4/IBIS/8/IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
13	**H7**	MASSA/NIMIR 3/3/YOGUI 1/TARASCA 87 3//HARE 212/4/IBIS/8/IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”

(continued)

Table 3.12.1. (continued)

No.	Name*	Pedigree
14	**JA**	IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”/8/35ITSN14
15	**H8**	MASSA/NIMIR 3/3/YOGUI 1/TARASCA 87 3//HARE 212/4/IBIS/8/IBIS/7/HARE 212/3/CHAMPLAIN/ARONDE 68//VPM/MOISSON/4/JUANILLO 100/5/ANDAS“S”/6/DURUM WHEAT/BALBO//BOK“S”/3/ANDAS“S”//TJ/BGL“S”
16	**BE**	29ITSN116/30ITSN34
17	**BF**	30ITYN36/29ITSN15
18	**Q1**	27ITYN45//12ITSN30/16ITSN64
19	**Q2**	27ITYN45//12ITSN30/16ITSN64
20	**X1**	W.TCL 83/HOHI//RHINO 4/3/ARDI 1/4/KIEWIET/5/A3165
21	**X2**	W.TCL 83/HOHI//RHINO 4/3/ARDI 1/4/KIEWIET/5/A3165
22	**X3**	W.TCL 83/HOHI//RHINO 4/3/ARDI 1/4/KIEWIET/5/A3165
23	**ZF**	22ITYN9/4/CHIVA“S”//YAR79/TH.JUNCEUM/3/17ITYN3/5/ 22ITSN41/16ITSN46-1-1//ROMANIA YIELD

* Codified due to confidentiality.

Chapter 4: Results and discussion

4.1. GENERAL AGRO-CLIMATIC CONDITIONS

The area of the Western Cape where the field trials were conducted (Figures 3.1.2 and 3.1.3) is located in a zone with a Mediterranean-type climate characterised by an extended summer period with little or no precipitation (especially the Swartland area), and a wet winter period when most rainfall occurs. Consequently, annual field crops under rainfed conditions are planted at the end of autumn (during the first half of May) and harvested at the end of spring (the first half of November). In such circumstances the triticale growth period normally spreads for a period of roughly 6 months, exposed to a short-day photoperiod (starting from 10h 20min in the middle of May to the shortest of 9h 53min during the last ten-day period (TDP) of June, with the longest days during the growth season being about 14h in the middle of November), and relatively low and moderate temperatures during the first half of growth season. The majority of cultivars and breeding lines are heading (show ear emergence) and begin grain filling during the second TDP of August, with only a few earlier maturing lines heading during its first TDP.

As can be seen from records of long-term precipitation and temperature data (Figures A4.1.1 – A4.1.20[**], Addendum 1) and HTC (Table 4.1.1), the general climatic regimen of the trial locations is characterised as dry (locations Piketberg, Vredenburg, Napier, Riversdale and Albertinia) and insufficiently wet (locations Klipheuwel, Langgewens, Roodebloem and Tygerhoek). The exception is Mariendahl location which is strongly affected by seasonal moist air masses coming from the North and North-West because of its proximity to a mountain range at the South-East (Figure 3.1.2-A). The mountain range causes distinctly different spatial and temporal precipitation patterns in the Swartland and Overberg regions during the growth season.

[**] The letter 'A' in front of the figure or table number stands for 'Addendum'.

Table 4.1.1. Selyaninov' hydro-thermal coefficient (HTC) for 2006 and 2007 seasons and long-term data

Location	TDP Mean	May			June			July			August			September			October			November		
		1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3
2006 season																						
VR	1.25		2.8	2.2	1.9	1.2	1.0	1.1	0.9	1.4	1.8	2.3	1.7	1.1	0.7	0.6	0.8	0.3	0.3	0.5		
LA	1.34		3.3	2.7	2.1	1.2	1.0	1.1	0.9	1.3	1.8	2.0	1.5	0.8	0.9	0.8	1.1	0.4	0.6	0.9	1.0	
ME	2.39	2.6	4.8	4.6	3.9	2.6	2.3	2.6	3.0	3.6	4.1	3.7	2.4	1.3	0.9	0.8	1.6	1.1	1.1	1.1	1.2	1.0
RO	1.17	0.9	1.6	1.2	1.1	0.7	0.8	0.7	1.0	1.9	2.4	2.3	1.7	1.4	1.2	0.6	0.9	0.6	0.6	0.9	1.0	
TY	1.11	0.8	1.3	1.1	1.1	0.6	0.7	0.4	1.5	2.4	2.9	2.1	2.2	1.8	1.5	0.2	0.2	0.2	0.3	0.6	0.5	
NA	1.30		0.8	1.1	1.0	0.9	0.4	0.4	1.9	3.5	3.7	2.6	1.8	1.8	1.4	0.5	0.7	0.3	0.3	0.3		
2007 season																						
PI	1.03	0.0	1.2	0.8	2.9	2.3	2.8	0.7	0.7	1.5	1.9	2.2	1.0	0.6	0.2	0.1	0.2	0.1	0.3	0.2		
LA	2.06		1.7	1.2	4.4	4.2	5.5	2.3	1.9	2.5	3.1	3.7	2.2	2.0	1.1	0.8	0.6	0.4	0.4	0.6	0.6	
KL	1.51		2.2	1.5	2.3	1.5	2.2	1.4	1.2	1.8	2.7	3.3	2.5	1.8	1.2	0.7	0.7	0.5	0.4	0.4	0.4	
ME	2.21	0.3	3.5	2.5	4.2	2.7	2.7	1.0	0.6	3.6	5.3	5.7	3.4	2.4	1.7	1.0	0.9	0.9	1.0	1.1	1.1	1.0
RO	1.27	0.2	1.3	1.1	1.3	0.9	2.4	2.4	2.2	1.9	2.2	2.4	1.3	0.9	0.6	0.2	0.8	0.7	1.0	0.4		
TY	0.57	0.0	0.8	0.7	0.8	0.4	0.7	0.6	0.7	1.1	1.1	1.1	0.3	0.3	0.2	0.2	0.4	0.4	0.6	0.4	0.6	
NA	0.92	0.0	0.8	0.7	1.0	0.5	0.9	1.1	1.1	2.1	2.1	2.1	0.9	0.6	0.6	0.4	0.8	0.7	0.7	0.2		
RI	0.93	0.1	2.1	2.1	2.1	1.0	0.8	0.8	0.9	1.4	1.4	1.4	0.6	0.6	0.3	0.3	0.5	0.5	0.5	0.2		
AL	0.93	0.0	1.6	2.0	2.0	1.1	0.8	0.8	0.8	1.0	1.2	1.3	0.6	0.5	0.3	0.3	0.9	1.0	1.1	0.3		
Long term data																						
PI	0.77	0.6	0.7	0.8	0.9	1.0	1.2	1.2	1.3	1.2	1.2	1.1	1.1	0.9	0.7	0.5	0.4	0.3	0.3	0.2	0.2	0.2
VR	0.91	0.6	0.4	0.7	0.9	1.2	1.2	1.4	1.6	1.6	1.5	1.4	1.4	1.3	1.1	0.8	0.5	0.5	0.4	0.4	0.3	0.2
LA	1.07	1.0	1.0	1.0	1.2	1.3	1.6	1.7	1.7	1.5	1.7	1.6	1.5	1.2	1.0	0.8	0.6	0.5	0.4	0.4	0.3	0.3
KL	1.22	0.4	1.6	1.3	1.6	1.1	1.3	1.2	1.3	1.9	2.5	2.9	2.2	1.4	0.8	0.6	0.8	0.6	0.5	0.5	0.5	0.6
ME	1.93	2.1	2.1	2.1	2.1	2.2	2.0	2.1	2.2	2.7	3.2	2.8	2.9	2.4	2.3	1.6	1.3	1.1	0.7	0.8	1.0	1.0
RO	1.04	1.0	0.9	1.0	1.1	1.2	1.3	1.4	1.5	1.4	1.4	1.5	1.6	1.4	1.1	0.7	0.7	0.7	0.6	0.5	0.4	0.3
TY	1.00	1.0	0.8	0.9	0.9	1.0	1.2	1.3	1.4	1.3	1.2	1.2	1.4	1.3	1.2	0.8	0.8	0.8	0.7	0.7	0.7	0.6
NA	0.92	0.8	0.7	0.8	0.9	0.9	1.1	1.3	1.4	1.4	1.2	1.2	1.2	1.1	1.0	0.7	0.6	0.7	0.6	0.6	0.6	0.5
RI	0.83	1.0	0.7	0.8	0.7	0.8	0.8	0.8	0.9	0.9	0.8	0.7	1.0	1.0	1.0	0.8	0.8	0.8	0.9	0.8	0.8	0.7
AL	1.06	0.2	0.8	0.9	1.2	1.2	1.1	0.9	0.8	1.3	1.2	1.2	1.1	1.1	0.9	0.4	0.8	1.0	1.1	0.6	1.5	2.9

Mariendahl is characterised by a generally very wet climatic pattern during the first 3/4 of the growth season. This leads to waterlogging of its shallow acidic sandy soils, which hinders proper development of a root system. As a result, plants with a weak root system are not prepared to withstand the following dry periods (if any) during grain filling.

In all locations most of the precipitation falls during the first half and middle (June-August) of the growth season. Frequent precipitation deficiency and rising temperatures in the last 1/3 of the growth season are interleaved with shorter periods of more favourable hydro-thermal conditions. Considering this, plants during grain filling period are usually exposed to a certain degree of precipitation deficit that results in lack of soil moisture. However, the soil type, its depth, plant root system development, and relative air humidity has to be taken into consideration at each particular location. Shortage of soil moisture during grain filling is often combined with high air temperatures and dry winds that cause conditions for atmospheric drought development.

4.2. AGRO-CLIMATIC CONDITIONS OF THE 2006 SEASON

The 2006 season was characterised by a generally higher level of total precipitation across all locations except Tygerhoek, where the precipitation did not differ much compared to the long-term data (Tables 4.2.1 and 4.1.1).

Table 4.2.1. Summary of the precipitation amounts for growth seasons (from planting to harvesting) across locations

Location / Season	PI	VR	LA	KL	ME	RO	TY	NA	RI	AL
Long term data, mm	216	248	294	322	582	284	280	241	236	252
2006 season, mm		289	339		689	323	278	310		
2007 season, mm	276		521	368	648	311	154	226	224	238

Temporal distribution of precipitation was not uniform with relatively short very wet periods interleaved with dry periods in the first part of the growth season and extended periods of dry spells during September-October across whole Overberg region, and at the Vredenburg in the Swartland region. Tygerhoek was the most affected by exposure to a dry spell throughout September-October. On the contrary, Mariendahl was over-wet during the first 2/3 of the season. The most potentially favourable weather conditions during the 2006 season were in Langgewens and Roodebloem.

4.3. AGRO-CLIMATIC CONDITIONS OF THE 2007 SEASON

The 2007 season in the Swartland region was generally similar to the 2006 season characterised by higher (compared to long-term data) amounts of precipitation with their favourable temporal distribution across all locations except Piketberg, where a dry spell was observed throughout the grain filling period from the last TDP of August until the harvest at the beginning of November. Conditions in the Overberg region were characterised by a generally lower (compared to long-term data) amount of total precipitation (except for Roodebloem) with an earlier start of dry spells (in August) during the second part of the growth season. In Tygerhoek a prolonged dry spell was observed from the first TDP of August until the middle of October. Trials across all locations (except Piketberg) received some good rains during the first TDP of October, which favoured grain filling of triticale lines with a longer growth period. Generally, the most potentially favourable weather conditions during the 2007 season were in Klipheuwel, Langgewens, Roodebloem, Napier and Albertinia, and the most unfavourable were in Tygerhoek, Piketberg and Mariendahl.

4.4. FIELD DATA ANALYSIS, 2006 AND 2007 SEASONS

Field *in situ* observations for plant height (cm) and days from planting to heading, as well as grain yield ($kg.ha^{-1}$) and test weight ($kg.HL^{-1}$) raw data are presented in Tables A4.4.1 – A4.4.15 (Addendum 2), and the scoring of lodging and adult plant resistance to rusts in Tables 4.4.16 and 4.4.17. In the 2007 season only rust-resistant lines (except for some check cultivars) were selected for planting, thus only few of them were affected by stem or leaf rusts compared to the 2006 season.

Grain yield data was subjected to a spatial analysis by means of a first-order autoregressive correlation model. Results of the spatial analysis for grain yield for each location trial of the 2006 and 2007 seasons are presented in Addendum 3. Plotted residual values are shown in Tables A4.4.18 – A4.4.36 (Addendum 4), and the summary of balanced least squares mean fixed values of grain yield are presented in Tables 4.4.37 – 4.4.39. The values of grain yield presented in Tables 4.4.37 – 4.4.39 were used for calculations of starch yield and ethanol yield (see below). As can be concluded from the residual values (Tables A4.4.18 – A4.4.36, Addendum 4), application of the spatial analysis was beneficial because of uneven field trial soil fertility (which incorporates many variables); the method was able to fix spatial differences of plots between and within blocks (replications). All the subsequent calculations where grain yield data was used were done on the basis of these fixed for spatial differences values. In addition, grain moisture content was determined and subtracted, thus all the values were calculated and reported on a dry weight basis (DWB), if not mentioned specifically otherwise (‘as-is’).

Table 4.4.16. Scoring of lodging, resistance to leaf and stem rusts of 2006 triticale elite trials (locations not affected by lodging or diseases are not shown)

No.	Name	Langgewens			Roodebloem			Tygerhoek			Napier		
		Lodging, %	Leaf rust	Stem rust	Lodging, %	Leaf rust	Stem rust	Lodging, %	Leaf rust	Stem rust	Lodging, %	Leaf rust	Stem rust
1	**CA**	0	40S	20S	0	S	20S	0	50S	30S	0	0	40MS-S
2	**CB**	0	30S	0	15	S	5MS	25	5S	5S, tS	0	100S	20MS-S
3	**CC**	80	5MS	0	0	MS	5MR	10	R-10S	tMS	0	0	25MR-MS
4	**CD**	0	0	90S	0	10MS-S	100S	0	10MS	80S	0	0	100S
5	**CE**	0	5MR-MS	5MR	10	5MR-MS	10MS-tMS	30	0	10MS, tS	20	0	20MR-tS
6	**D1**	80	0	0	50	0	R	40	0	0	25	R	R
7	**DB**	0	0	0	20	tR-MR	10MS-tS	40	0	tS	0	0	30MR-5S
8	**YA**	0	0	5MS-S	20	0	40MS-S, 5S	10	0	30MS-S, tS	0	0	30MS-S
9	**DC**	0	5MS	0	60	R	5MR, tS	10	0	5MS	0	0	R-tMS
10	**YB**	80	10S	10S	20	tMS	10MS-S, tS	0	0	10MS, tMS	10	80S	20MS-S
11	**YC**	0	0	10S	20	tMS	10MS-S, tS	0	0	10S, tS	0	0	30MR-5S
12	**DD**	0	0	R	60	0	20MR-MS	20	0	5MS-S, tS	0	0	40MR
13	**DE**	0	0	0	50	0	tMS, tR	0	0	10MS-S, tS	0	0	20MR-tS
14	**DF**	80	0	0	30	0	10MR-S	0	0	5MS-S	0	0	30S
15	**DG**	80	0	20S	40	5MS	40MS-S, 5S	35	0	30S	0	0	40S
16	**DH**	0	0	10S	60	0	60S, 20S	20	0	50S	20	0	40S
17	**Y1**	0	0	tMS-tS	25	tR-MS	20S, tR-tMS	0	0	25S	25	0	20MR-MS
18	**Y2**	0	0	tMS	20	0	20S, 5S	0	0	20S, tS	0	0	30MR-MS
19	**YD**	0	0	tMS-tS	25	0	20S, tR-5S	0	0	20MS-S, tS	25	0	30MS-S
20	**Y3**	0	0	tR	20	0	20MS-S, 5MS-S	0	0	30S, tS	0	0	20MS-S

Table 4.4.17. Scoring of lodging, resistance to leaf and stem rusts of 2007 triticale elite trials (trials not affected by lodging or diseases are not shown)

No.	Name	Mariendahl			Langgewens			Riversdale			Albertinia		
		Lodging, %	Leaf rust	Stem rust	Lodging, %	Leaf rust	Stem rust	Lodging, %	Leaf rust	Stem rust	Lodging, %	Leaf rust	Stem rust
1	**CA**	0	10MS	10S	0	0	40MS	0	0	0	0	0	0
2	**CC**	0	5MS	0	0	0	60MS	0	0	0	0	0	0
3	**CE**	0	5MS	5MS	0	0	30MS	0	0	0	0	0	0
4	**CD**	0	60MS	40MS	10	0	40MS	0	0	0	0	0	0
5	**D1**	0	0	0	0	0	0	0	0	0	5	0	0
6	**D2**	0	0	0	30	0	0	40	0	0	10	0	0
7	**D3**	0	0	0	0	0	0	0	0	0	10	0	0
8	**D4**	0	0	0	0	0	0	30	0	0	0	0	0
9	**YC**	0	0	0	0	0	0	0	0	0	0	0	0
10	**Y2**	0	0	0	0	0	0	0	0	0	0	0	0
11	**EA**	0	0	0	0	0	0	0	0	0	0	0	0
12	**EB**	0	0	0	0	0	0	0	0	0	0	0	0
13	**G1**	0	0	0	0	0	0	0	0	0	0	0	0
14	**G2**	0	0	0	0	0	0	0	0	0	0	0	0
15	**H1**	0	0	0	60	0	0	80	0	0	0	0	0
16	**H2**	0	0	0	60	0	0	40	0	0	0	0	0
17	**H3**	0	0	0	0	0	0	60	0	0	0	0	0
18	**H4**	0	0	0	0	0	0	0	0	0	0	0	0
19	**H5**	0	0	0	0	0	0	0	0	0	0	0	0
20	**H6**	0	0	0	0	0	0	0	0	0	0	0	0

Table 4.4.37. Grain yield balanced least squares mean fixed values (kg.ha^{-1}) of 2006 triticale elite trials corrected by spatial analysis

Entry	Line code name	Location						Mean	Standard deviation
		VR	LA	ME	RO	TY	NA		
1	**CA**	2979.28	3256.90	3064.63	6138.02	4190.47	5201.19	4138.42	1183.52
2	**CB**	3116.22	1917.47	1903.67	4779.56	4104.33	4378.61	3366.65	1145.11
3	**CC**	3370.76	2848.86	2512.22	5319.70	4309.74	5798.28	4026.59	1224.86
4	**CD**	3878.40	3288.04	2115.64	5562.08	2828.42	3938.51	3601.85	1075.32
5	**CE**	3012.61	2780.43	3018.69	5591.57	3984.38	4644.85	3838.75	1018.04
6	**D1**	2978.07	2539.14	2957.88	5369.56	3670.05	5446.13	3826.81	1166.21
7	**DB**	3089.92	2874.65	2775.35	5793.95	3830.68	5473.57	3973.02	1225.32
8	**YA**	3137.99	3486.64	2666.12	5568.33	4259.05	5325.38	4073.92	1082.93
9	**DC**	3129.18	3988.49	2990.98	4913.97	4081.83	5280.18	4064.11	839.92
10	**YB**	4321.33	3481.92	2636.08	6075.93	4668.43	3729.29	4152.16	1074.13
11	**YC**	3775.99	2961.40	2590.33	6037.18	4858.22	5767.86	4331.83	1320.82
12	**DD**	3544.12	2903.36	2703.27	5173.98	3766.46	4954.49	3840.95	938.44
13	**DE**	3041.00	3247.21	2950.99	5506.24	4241.38	5121.38	4018.03	1014.02
14	**DF**	3207.31	2994.69	2763.73	5376.27	4023.72	4806.06	3861.96	965.87
15	**DG**	3192.50	2861.81	2318.25	5201.52	3773.09	3914.16	3543.56	915.45
16	**DH**	2533.71	2985.03	2675.46	5885.83	3709.46	5217.81	3834.55	1284.14
17	**Y1**	2539.44	3397.93	2784.43	5964.63	4281.88	5762.16	4121.75	1349.62
18	**Y2**	2808.72	3098.76	2716.56	5736.43	4647.86	6138.85	4191.20	1394.56
19	**YD**	2730.74	3677.87	2480.03	5775.12	4294.75	5180.77	4023.21	1200.97
20	**Y3**	2742.05	3259.26	3345.50	5699.17	4411.08	6052.34	4251.57	1254.86
	Mean (site index)	3156.47	3092.49	2698.49	5573.45	4096.76	5106.59		
	Standard deviation	436.40	429.02	324.45	366.16	431.72	678.25		

Table 4.4.38. Grain yield balanced least squares mean fixed values (kg.ha^{-1}) of the 2006 Mariendahl triticale senior trials

Entry	Block MA		Block MB		Block MC		Block MD		Mean	Standard deviation
	Line code name	Grain yield	Line code name	Grain yield	Line code name	Grain yield	Line code name	Grain yield		
1	**CA**	3132.81	**CA**	3075.16	**CA**	2626.50	**CA**	2634.89	2867.34	237.54
2	**CB**	1766.09	**CB**	1902.83	**CB**	1760.31	**CB**	2236.02	1916.31	193.19
3	**CC**	2601.94	**CC**	2673.15	**CC**	2250.38	**CC**	2282.01	2451.87	187.71
4	**CD**	1520.19	**CD**	1711.37	**CD**	1708.96	**CD**	1934.15	1718.67	146.60
5	**CE**	2872.20	**CE**	2450.86	**CE**	2329.59	**CE**	2480.54	2533.30	203.67
6	**BA**	2453.54	**BP**	2491.06	**UD**	2534.03	**UT**	2520.08		
7	**BB**	2301.65	**BQ**	2345.28	**UE**	2358.27	**UU**	2586.13		
8	**BC**	2770.28	**BR**	2361.34	**UF**	2060.12	**UV**	2588.08		
9	**BD**	2341.21	**BS**	2009.52	**UG**	2766.99	**UW**	2845.56		
10	**BE**	2780.97	**BT**	2628.99	**UH**	2284.82	**UX**	2478.60		
11	**BF**	2527.63	**BU**	2156.54	**UI**	1771.28	**UY**	2682.53		
12	**BG**	2443.76	**BV**	2507.37	**UJ**	2841.35	**UZ**	2769.45		
13	**BH**	2692.39	**BW**	2509.87	**UK**	2570.72	**ZA**	2776.98		
14	**BI**	3045.58	**BX**	2075.56	**UM**	1936.18	**ZB**	2810.76		
15	**BJ**	3061.30	**BY**	2257.38	**UN**	2323.47	**ZC**	2822.78		
16	**BK**	2429.32	**BZ**	2409.78	**UO**	2669.94	**ZD**	2831.15		
17	**BL**	2440.67	**UA**	2450.57	**UP**	2618.01	**ZE**	1898.01		
18	**BM**	2413.36	**EA**	2717.85	**UQ**	2230.11	**ZF**	1797.71		
19	**BN**	3124.72	**UB**	2589.52	**UR**	1995.02	**ZG**	2153.42		
20	**BO**	2975.73	**UC**	2376.98	**US**	2101.50	**ZH**	2309.83		

Table 4.4.39. Grain yield balanced least squares mean fixed values (kg.ha^{-1}) of the 2007 triticale elite trials corrected by spatial analysis

Entry	Line code name	Location									Mean	St. Dev.
		PI	LA	KL	ME	RO	TY	NA	RI	AL		
1	**CA**	5174.99	5996.79	4897.89	3349.67	7220.78	5587.40	6180.08	3689.86	5291.16	5265.40	1135.05
2	**CC**	4468.97	5447.02	4169.03	3686.90	6980.35	5599.86	6042.99	5675.22	6913.18	5442.62	1086.23
3	**CE**	4992.40	4864.09	4718.28	3837.12	5869.00	5323.22	5069.24	6068.22	5944.92	5187.39	669.37
4	**CD**	5170.71	4675.65	5508.17	1947.52	6326.53	3728.22	648.97	2069.40	4449.25	3836.05	1786.29
5	**D1**	5191.21	6257.32	4866.52	4432.89	6819.85	6091.06	5813.40	5287.09	6339.78	5677.68	735.22
6	**D2**	4990.40	5931.31	5058.07	4136.73	6978.74	5546.43	6020.26	6378.62	6669.67	5745.58	853.51
7	**D3**	5058.36	5431.30	4158.56	3491.71	7363.32	5838.39	6197.99	5710.72	6084.88	5481.69	1079.59
8	**D4**	4826.04	5666.67	5064.62	3526.38	7492.66	5807.93	6196.06	5973.28	6774.15	5703.09	1084.90
9	**YC**	5216.60	5476.48	5009.43	3086.71	7199.72	6279.99	5610.03	5886.18	6667.09	5603.58	1109.04
10	**Y2**	4946.67	5998.70	4569.58	4003.79	5648.92	6078.06	6469.40	4660.67	6592.28	5440.90	870.59
11	**EA**	4670.21	6502.37	4578.60	4297.37	5889.43	6040.52	5387.86	5470.54	6769.25	5511.79	820.27
12	**EB**	4159.80	5324.98	4240.28	3261.55	5513.43	5113.58	4741.05	4946.12	5699.75	4777.84	730.51
13	**G1**	5193.94	6341.55	4234.34	3344.01	7447.38	5754.29	5889.58	5621.63	6782.01	5623.19	1182.40
14	**G2**	5732.55	6106.95	3925.25	3759.36	7924.82	6348.88	6003.20	5791.14	7091.27	5853.71	1259.48
15	**H1**	5237.46	6278.76	4339.32	3628.17	5827.49	5460.87	6595.82	5669.87	6947.55	5553.92	996.33
16	**H2**	5518.57	5497.38	4972.88	3237.88	6325.89	5452.39	6479.19	5555.84	6397.31	5493.04	933.72
17	**H3**	5028.63	6064.15	4902.78	4057.19	6367.59	6444.51	5615.34	5443.72	6554.25	5608.69	791.99
18	**H4**	5006.07	5584.45	4904.50	2846.46	5663.46	5795.87	4898.80	6074.55	5944.21	5190.93	930.38
19	**H5**	4995.46	4829.03	4080.93	3071.00	6261.51	4062.81	3874.13	4631.75	5344.97	4572.40	878.21
20	**H6**	5146.60	5433.81	4357.80	3233.88	6874.93	6508.98	6462.32	5347.98	5955.71	5480.22	1086.84
Mean (site index)		5036.28	5685.44	4627.84	3511.81	6599.79	5643.16	5509.79	5297.62	6260.63		
Standard Deviation		329.56	509.19	412.08	554.14	704.51	691.93	1301.22	950.49	657.01		

4.5. MOISTURE AND TOTAL STARCH ANALYSES, THE 2006 AND 2007 SEASONS

Results of moisture (%) and total starch (% DWB) content analyses are presented in Tables 4.5.2 and 4.5.3. Cross-site analysis of total starch content was done for the 2007 season data (Addendum 5) which revealed that lines H6, G1, H3 and D4 were the best in terms of total starch content and its stability across locations.

Analysis of variance (ANOVA) of the 2007 moisture and starch contents for the Mariendahl trial' three repetitions (see Table 4.5.3) are presented in Table 4.5.1. For both moisture and starch, differences in variance between the field trial repetitions were significant at 5% significance level. Therefore, it could be recommended that sub-samples should be taken from each repetition and mixed into a composite sample prior to analyses in order to obtain an average values per entry.

Table 4.5.1. ANOVA (single factor) of moisture and starch contents for three replications of the 2007 Mariendahl triticale elite trial

(A) Moisture content

Repetition	Count	Sum	Average	Variance
1	20	207.19	10.3595	0.077721
2	20	202.78	10.1390	0.035494
3	20	203.08	10.1540	0.107520

Moisture content - ANOVA

Source of Variation	SS	df	MS	F	P-value	F critical
Between rep's	0.607170	2	0.303585	4.126021	0.021208	3.158843
Within rep's	4.193955	57	0.073578			
Total	4.801125	59				

(B) Starch content 'as-is'

Repetition	Count	Sum	Average	Variance
1	20	1188.506	59.42532	2.103311
2	20	1184.597	59.22983	1.972515
3	20	1115.442	55.77212	4.480177

Starch content 'as is' - ANOVA

Source of Variation	SS	df	MS	F	P-value	F critical
Between rep's	168.9323	2	84.46615	29.61645	1.52E-09	3.158843
Within rep's	162.5641	57	2.852001			
Total	331.4964	59				

Table 4.5.2. Contents of moisture (Mo., %) and total starch (St., % dry matter) in the 2006 triticale elite and senior trials

No.	Location / Name	ME		LA		VR		RO		TY		NA		MA		MB		MC		MD	
		Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.
1	**CA**	10.75	59.88	10.77	69.40	11.21	67.53	10.98	65.46	11.81	60.91	11.31	67.29	10.34	62.27	11.47	63.17	10.71	62.23	11.12	62.88
2	**CB**	10.81	55.71	10.24	66.95	10.91	64.58	11.28	64.84	11.42	60.18	11.03	64.38	9.77	59.37	10.71	58.63	10.78	59.18	10.38	57.39
3	**CC**	10.69	58.53	10.50	62.70	10.98	62.66	10.87	68.08	11.27	60.17	10.91	66.06	10.01	61.02	11.10	60.26	10.75	63.32	10.77	63.62
4	**CD**	10.95	61.61	10.38	68.11	10.96	63.31	11.19	65.05	11.48	62.55	11.19	68.60	10.82	62.34	10.80	61.64	10.87	63.18	11.04	66.55
5	**CE**	10.74	62.96	10.41	64.41	10.87	62.02	11.08	65.53	11.35	60.13	10.85	65.08	10.72	62.64	10.35	62.22	10.98	62.66	11.37	64.87
6	**D1**	10.04	64.10	10.79	61.09	10.62	62.74	11.09	65.91	11.54	60.13	11.32	59.75	10.70	61.90	10.79	62.57	10.76	62.62	11.38	63.62
7	**DB**	10.27	63.62	10.48	57.95	10.48	64.61	11.01	65.29	11.09	61.71	11.12	59.05	10.48	61.38	10.78	63.03	10.75	61.56	11.29	62.80
8	**YA**	10.32	63.18	10.45	61.62	10.41	61.62	10.81	64.22	11.31	62.43	11.13	61.90	10.39	62.50	11.19	63.13	10.60	59.26	11.33	63.04
9	**DC**	10.14	60.27	10.48	61.26	10.19	64.40	11.06	64.40	11.05	61.77	11.37	58.65	10.56	62.25	10.67	60.98	11.22	60.35	11.24	64.06
10	**YB**	10.35	62.65	10.29	60.37	10.18	63.08	10.59	61.37	10.96	61.24	11.07	56.94	10.47	64.90	9.51	58.45	10.65	60.92	11.20	62.77
11	**YC**	10.51	60.70	10.35	57.86	10.14	61.84	11.48	65.28	10.87	60.81	11.61	55.95	10.28	61.24	10.97	63.63	10.52	60.83	10.83	62.96
12	**DD**	10.11	62.85	10.41	59.89	10.75	63.58	11.10	63.96	10.88	61.76	11.81	60.47	10.68	63.70	10.67	58.08	10.84	61.05	11.42	64.64
13	**DE**	10.02	60.74	10.32	59.92	10.38	61.26	11.52	63.22	11.10	60.69	11.50	59.69	10.21	59.66	10.70	58.57	10.88	61.46	11.44	62.30
14	**DF**	10.07	62.54	10.38	58.36	10.21	61.33	11.36	64.24	11.16	65.41	11.14	59.35	10.57	64.34	10.81	62.02	10.65	60.73	11.59	64.95
15	**DG**	10.23	61.17	9.96	59.31	10.04	61.49	11.32	62.32	10.94	62.83	10.87	60.67	10.96	62.26	10.66	61.26	10.78	62.64	11.02	63.89
16	**DH**	9.80	59.95	9.71	61.11	10.14	62.40	11.54	65.13	11.18	64.84	11.03	61.45	10.17	59.91	10.77	60.58	10.05	61.46	11.21	63.65
17	**Y1**	10.00	60.55	10.07	62.73	10.39	62.01	10.90	63.06	11.02	64.27	11.32	58.78	10.03	58.19	10.91	62.65	10.48	59.94	11.08	63.84
18	**Y2**	9.92	61.05	10.44	62.52	10.54	63.06	11.27	58.96	11.05	64.52	11.34	56.60	10.68	62.34	10.91	65.01	10.55	61.62	10.90	64.63
19	**YD**	9.98	62.02	10.39	59.86	10.49	61.99	11.24	56.48	10.65	66.15	11.44	63.07	10.75	61.20	10.98	60.64	10.27	60.85	11.26	66.38
20	**Y3**	10.30	62.52	10.24	63.69	10.33	61.23	10.77	61.27	11.25	64.44	11.06	62.99	10.35	63.64	10.62	62.17	10.15	60.48	11.00	65.07

Table 4.5.3. Contents of moisture (Mo., %) and total starch (St., % dry matter) in the 2007 triticale elite trials

No.	Location-rep.	ME-1		ME-2		ME-3		LA		PI		KL		RO		TY		NA		RI		AL	
	Name	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.	Mo.	St.
1	CA	11.17	66.09	10.29	67.49	10.20	63.90	11.51	66.17	10.85	67.56	11.34	61.31	11.60	64.05	11.05	62.85	11.03	62.69	10.83	59.59	10.98	61.82
2	CC	10.51	65.12	10.49	65.56	10.10	59.46	10.91	65.13	10.44	64.99	10.82	65.67	10.97	62.24	10.93	59.92	10.65	63.43	10.28	59.81	11.26	60.34
3	CE	10.28	64.65	10.21	65.03	10.11	62.48	10.68	63.95	10.22	66.13	11.31	63.54	10.70	67.14	11.05	61.64	10.76	63.78	10.84	61.09	11.04	58.73
4	CD	10.40	64.53	10.18	63.14	10.37	60.31	11.10	64.36	10.42	66.33	11.15	63.76	10.69	66.44	11.10	63.08	10.75	62.62	10.30	59.00	10.90	62.53
5	D1	10.02	65.02	10.46	63.88	10.16	62.22	11.27	64.14	10.76	67.39	11.25	63.00	11.00	64.93	10.92	62.77	10.87	63.82	10.75	60.54	11.24	62.03
6	D2	10.00	66.00	10.13	65.73	10.01	63.61	11.15	64.35	10.56	65.00	11.04	64.67	10.83	67.10	10.72	62.45	10.92	64.58	10.95	62.21	11.06	61.62
7	D3	10.36	67.83	10.07	67.12	9.77	59.85	10.69	64.90	10.91	65.42	11.01	64.64	10.99	66.57	10.52	62.21	10.87	67.46	10.94	63.45	11.31	62.14
8	D4	10.10	65.48	10.08	65.31	9.92	61.67	11.18	65.08	10.78	65.11	11.15	65.52	10.75	67.27	10.57	62.51	10.82	67.43	10.70	59.26	11.49	61.28
9	YC	9.93	62.54	9.92	63.65	10.52	57.87	10.79	64.27	10.70	66.35	11.21	62.09	11.01	64.27	10.87	60.56	10.68	64.68	10.47	58.51	11.36	60.41
10	Y2	10.26	64.99	10.38	67.57	11.00	62.31	11.08	66.30	10.56	65.01	10.93	61.86	10.93	65.13	10.69	61.66	11.22	63.37	10.79	61.72	11.62	60.23
11	EA	10.13	67.37	10.26	66.70	10.71	65.79	11.14	65.95	10.93	66.15	10.95	65.17	10.79	65.96	10.34	61.03	10.97	65.12	10.62	59.56	11.07	59.76
12	EB	10.36	67.65	10.21	64.15	10.19	59.78	11.68	64.84	10.77	61.98	10.84	64.89	10.96	65.12	11.15	58.79	10.68	63.28	10.67	60.22	11.63	60.27
13	G1	10.34	67.75	9.86	68.64	10.46	64.05	11.31	66.35	10.79	67.87	10.83	65.73	11.01	67.23	11.17	63.04	10.93	66.00	10.56	61.63	11.53	63.30
14	G2	10.40	66.21	9.77	64.97	10.18	63.50	11.21	66.04	10.57	66.89	10.87	63.79	10.13	64.62	11.18	62.52	10.90	65.21	10.50	62.38	11.78	63.88
15	H1	10.35	67.29	10.01	66.52	9.92	64.02	11.38	66.19	10.66	66.63	10.93	63.69	11.09	65.34	11.30	62.65	11.06	62.01	10.74	59.72	11.18	62.19
16	H2	10.50	66.98	10.16	67.75	10.02	60.41	10.75	66.07	10.88	65.21	10.64	63.82	10.48	65.10	10.99	63.39	10.78	65.41	10.37	61.41	11.24	62.66
17	H3	10.41	70.31	10.21	67.13	9.97	61.04	11.43	67.13	11.12	67.74	10.92	64.17	10.61	66.00	11.55	64.73	10.64	63.93	10.35	64.36	12.02	63.23
18	H4	10.80	66.61	9.95	65.30	9.97	61.17	10.93	65.11	10.96	64.68	10.84	65.52	10.60	63.96	11.32	62.13	10.62	66.46	10.25	61.45	11.73	62.11
19	H5	10.42	65.88	10.03	65.08	9.90	60.60	11.36	66.05	10.88	67.05	11.02	65.27	10.66	65.13	10.99	59.68	10.68	62.00	10.22	59.06	11.92	61.68
20	H6	10.45	67.59	10.11	67.53	9.60	67.47	10.84	66.56	11.10	66.65	10.70	65.52	10.80	65.03	11.25	62.11	10.60	66.67	10.56	65.76	11.46	64.06

In 2007, three stable commercial cultivars-checks (CA, CC and CE) from the second repetition of the Mariendahl trial were assessed for moisture content by the moisture scale method, and by the mechanical-convection air-oven method (modified AACC International Method 44-15.02). Three repetitions were done for each entry. The results are compared in Table 4.5.4.

Table 4.5.4. Comparison of moisture content (%) results analysed by moisture scale and drying oven methods

Sample - repetition	Moisture scale, 3g sample at 110°C with automatic shut-down	Drying oven, 5g sample at 130°C			
		2h	3h	4h	20.5h
CA - 1	10.50	12.61	12.81	12.67	13.17
CA - 2	10.71	12.48	12.74	12.62	13.04
CA - 3	10.75	12.62	12.78	12.70	13.24
Average	10.65	12.57	12.78	12.66	13.15
Difference between repetitions*	±1.17%	±0.56%	±0.26%	±0.32%	±0.76%
Variance	0.018033	0.00599			
CC - 1	10.30	12.40	12.62	12.50	13.04
CC - 2	10.63	12.36	12.56	12.46	13.00
CC - 3	10.47	12.35	12.47	12.45	13.07
Average	10.47	12.37	12.55	12.47	13.04
Difference between repetitions*	±1.58%	±0.19%	±0.59%	±0.19%	±0.27%
Variance	0.027233	0.00062			
CE - 1	10.69	12.37	12.53	12.49	13.03
CE - 2	10.32	12.45	12.61	12.57	13.09
CE - 3	10.52	12.36	12.44	12.48	13.02
Average	10.51	12.39	12.53	12.51	13.05
Difference between repetitions*	±1.76%	±0.34%	±0.66%	±0.34%	±0.25%
Variance	0.034300	0.00215			

ANOVA: Two-Factor with replication for moisture scale and drying oven (2h) methods

Source of Variation	SS	df	MS	F	P-value	F critical
Cultivars	0.128265	2	0.064132	4.356246	0.037818	3.885294
Drying methods	16.288480	1	16.288480	1106.41	3.46E-13	4.747225
Interaction	0.000881	2	0.000440	0.029912	0.970603	3.885294
Within	0.176663	12	0.014722			
Total	16.594280	17				

* Values of the difference between repetitions were calculated according to the formula: ((***Max***(repetition) – ***Min***(repetition)) / Average) × (100 / 2)

As can be seen, standard drying-oven method with 2h drying time gives *ca*. 2% higher values for moisture content (compare averages from columns 2 and 3), and also gives more stable results across repetitions (considering difference between repetitions and variance) compared to the express moisture scale method. This was true for all three cultivars analysed. Thus, some moisture remains unaccounted for if moisture scale method is used, with the error conveyed to the following calculations. It would be advisable to use standard AACC International Method 44-15.02 for moisture determination in future research.

4.6. PROTEIN AND PSI DETERMINATION, THE 2007 SEASON

Protein content in whole grain (% at 12% moisture basis) and particle size index (PSI, %) for the 2007 season are shown in Table 4.6.1 (protein and PSI raw data courtesy of Dr MARENA MANLEY group, Department of Food Science, Stellenbosch University, RSA, 2009). Higher PSI values are indicative of softer grain (DU PISANI, 2009). Protein data had some extreme values (below 5%) which might be due to outliers or error of the method employed. Thus, protein data has to be taken into consideration cautiously. A standard method for protein determination is advisable for more accurate results.

Table 4.6.1. Protein content (Pr., % dry matter) and particle size index (PSI, %) in the 2007 triticale elite trials

		Swartland																					
	Location-rep.	ME-1		ME-2		ME-3		LA-1		LA-2		LA-3		PI-1		PI-2		PI-3		KL-1		KL-2		KL-3	
No.	Name	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI
1	CA	8.9	71.4	6.8	75.6	5.5	75.9	9.2	71.9	7.2	73.5	10.9	68.0	7.0	73.7	7.6	73.8	6.9	76.3	11.7	68.6	11.7	66.8	11.1	72.2
2	CC	6.0	65.2	4.8	64.9	4.4	75.9	9.1	63.5	9.6	67.4	8.6	70.0	8.0	71.5	8.5	69.5	7.4	69.4	12.2	66.3	11.5	67.9	10.0	66.8
3	CE	8.3	66.9	5.9	67.6	8.1	67.2	8.8	68.1	11.5	64.6	10.1	63.1	7.5	65.4	8.6	65.6	6.8	66.5	13.9	64.0	12.8	63.1	12.3	64.2
4	CD	7.0	68.7	5.7	69.6	5.6	69.8	9.3	67.0	9.6	65.9	10.5	67.9	7.3	66.9	8.1	70.9	6.6	70.5	11.3	67.4	9.0	67.7	9.6	64.2
5	D1	6.2	70.1	6.9	72.0	5.0	68.8	9.9	64.5	10.8	67.2	11.8	64.5	6.9	68.2	9.0	67.7	6.6	70.2	12.7	66.3	12.0	67.0	12.5	65.5
6	D2	6.8	70.5	7.1	70.0	5.5	69.4	9.4	66.0	9.9	67.5	11.5	64.0	7.2	65.9	7.6	69.6	7.1	69.0	13.2	64.2	10.7	65.2	11.5	63.7
7	D3	7.4	67.8	5.6	65.6	4.7	71.8	11.3	65.3	9.1	68.0	12.4	64.4	7.0	70.1	8.4	68.1	6.5	72.2	13.7	64.7	12.4	63.9	12.2	64.9
8	D4	6.7	68.0	7.1	70.1	5.7	73.1	10.7	65.2	9.0	69.2	10.7	68.2	7.5	67.8	7.6	70.6	7.6	68.9	13.2	64.0	11.4	64.8	12.6	66.6
9	YC	6.9	73.9	6.4	73.8	4.3	75.7	11.0	64.3	9.7	66.5	9.9	68.9	7.2	70.0	7.5	72.4	7.9	70.2	13.6	65.1	11.3	67.7	11.5	66.9
10	Y2	8.5	72.5	5.8	77.6	6.7	76.6	9.9	68.5	9.4	70.2	13.3	64.1	6.9	74.8	8.8	72.1	8.1	71.5	12.3	65.0	13.0	67.7	11.1	69.2
11	EA	7.7	72.4	7.0	68.9	6.0	69.8	9.9	68.0	8.2	70.6	11.6	67.1	7.7	70.2	8.2	70.6	8.4	69.4	14.7	62.7	13.1	65.9	12.3	64.6
12	EB	8.9	72.6	9.6	69.5	6.4	70.4	13.0	63.3	12.3	64.3	14.2	62.9	9.9	66.9	10.2	65.3	9.7	67.9	15.3	63.0	14.6	63.6	12.2	62.6
13	G1	6.2	72.9	5.6	69.6	5.8	68.9	9.8	65.1	8.2	65.1	9.7	69.7	7.2	70.2	7.8	71.4	9.7	70.5	12.7	64.7	10.9	65.5	11.5	65.5
14	G2	7.6	69.9	6.9	73.7	6.2	70.1	10.3	65.5	7.4	65.7	8.8	68.5	7.9	69.6	8.6	69.2	8.7	72.4	12.6	63.5	10.2	66.5	10.6	66.6
15	H1	6.6	68.2	5.6	70.3	5.8	70.7	10.3	66.0	8.8	69.1	11.6	65.9	7.5	66.4	8.0	71.2	7.3	70.6	13.2	68.0	10.9	65.8	10.0	69.1
16	H2	5.3	71.5	5.6	68.4	4.8	69.9	11.8	63.8	8.0	68.7	11.1	66.1	7.2	67.7	7.9	69.8	7.3	72.0	12.8	66.1	11.4	65.1	11.3	66.9
17	H3	6.7	70.8	5.7	67.8	5.1	74.4	10.7	66.3	8.9	69.4	10.1	69.6	6.9	70.7	7.8	71.5	7.9	70.0	12.6	65.3	10.9	65.2	12.7	63.1
18	H4	6.5	70.8	7.2	68.4	5.1	68.5	9.7	67.0	9.7	67.9	10.2	67.9	8.7	67.7	9.1	66.7	9.8	68.7	12.6	64.7	11.5	65.7	11.0	67.8
19	H5	7.6	72.6	6.0	67.3	6.9	66.7	12.1	66.1	9.3	65.5	10.7	66.9	10.1	64.1	9.3	65.7	9.5	68.0	13.6	63.7	12.5	63.3	11.1	64.6
20	H6	7.2	71.4	6.7	68.8	6.3	72.8	10.7	65.5	10.8	67.1	10.4	66.7	7.0	73.3	8.6	70.8	8.4	73.4	14.4	62.7	12.7	64.5	13.0	65.6

(continued)

Table 4.6.1. (continued)

Overberg (Ruens)																															
	Location-rep.	RO-1		RO-2		RO-3		TY-1		TY-2		TY-3		NA-1		NA-2		NA-3		RI-1		RI-2		RI-3		AL-1		AL-2		AL-3	
No.	Name	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI	Pr.	PSI
1	**CA**	6.8	67.6	10.2	66.5	9.4	67.6	11.7	61.7	11.7	60.7	11.5	61.1	13.9	66.1	10.0	67.8	11.9	65.7	10.9	65.2	10.9	65.3	11.0	63.6	15.4	63.1	11.2	61.3	11.4	61.9
2	**CC**	8.7	64.8	10.6	59.9	10.5	62.2	11.3	59.3	11.6	56.8	11.3	58.3	13.0	61.8	11.8	62.9	14.2	61.3	10.8	58.5	10.8	60.9	11.1	60.7	15.6	64.1	11.1	58.7	11.1	59.8
3	**CE**	8.6	63.9	11.3	61.3	10.6	62.9	11.6	57.0	11.4	59.1	11.3	58.0	12.6	61.0	13.8	58.0	14.1	58.0	11.0	56.9	10.9	59.9	11.0	59.9	16.7	60.2	11.2	58.5	11.2	58.5
4	**CD**	10.0	66.1	11.0	65.3	11.2	66.1	11.3	60.6	11.5	60.2	11.9	57.9	11.4	64.4	11.2	62.7	13.8	59.6	11.0	60.8	11.1	58.0	11.0	60.5	15.0	62.9	11.4	59.0	11.5	56.8
5	**D1**	9.9	65.4	9.9	64.1	9.9	66.5	11.6	57.3	10.9	62.1	11.7	56.6	14.5	60.3	9.8	67.3	14.4	61.3	10.8	63.2	11.1	58.8	11.2	60.3	13.7	63.1	11.2	61.2	11.4	62.0
6	**D2**	9.3	61.9	10.3	63.5	10.0	63.9	11.2	61.1	11.9	58.4	11.5	59.8	14.7	60.5	12.9	60.2	15.5	59.5	11.0	60.7	11.1	60.0	11.1	62.1	15.1	61.5	11.2	59.9	11.3	57.7
7	**D3**	9.5	65.2	12.0	62.5	8.1	67.1	11.6	57.4	11.3	57.6	11.7	57.5	13.7	64.5	12.5	67.3	12.1	61.8	11.0	59.2	11.1	62.1	11.1	61.2	14.9	61.7	11.3	61.3	11.5	56.8
8	**D4**	9.6	65.3	10.1	65.2	10.2	61.2	11.5	56.9	11.1	56.6	11.3	57.3	15.0	62.1	14.1	64.3	13.2	61.8	10.8	60.2	11.0	59.2	11.0	59.9	13.1	64.5	11.2	60.1	11.3	59.1
9	**YC**	10.2	67.6	9.5	68.8	10.0	67.2	12.7	64.0	11.7	60.2	11.6	62.6	15.2	61.3	13.5	63.0	13.1	63.1	11.1	58.1	11.0	65.6	10.9	68.4	14.9	58.7	11.4	63.3	11.1	62.4
10	**Y2**	9.1	70.2	13.0	64.3	10.7	64.7	12.1	62.6	11.4	61.3	11.4	63.3	15.6	65.4	11.7	67.6	14.8	63.0	10.9	64.2	11.1	64.0	11.0	67.8	16.3	63.4	11.2	63.2	11.1	64.3
11	**EA**	11.0	68.3	12.8	61.7	11.3	66.2	11.0	62.6	11.1	62.2	11.4	57.9	15.2	57.9	13.5	61.7	16.4	59.0	10.9	62.8	11.2	58.0	10.9	65.6	15.2	63.3	11.2	60.6	11.4	57.3
12	**EB**	12.9	64.3	12.9	59.2	11.2	61.6	11.5	59.8	11.6	56.5	11.5	57.7	16.0	61.9	14.1	63.8	13.9	66.8	10.7	65.7	11.0	61.3	11.2	60.7	16.3	60.4	11.3	60.7	11.6	56.2
13	**G1**	10.8	65.1	10.4	63.6	11.1	64.4	11.8	59.9	11.4	62.2	11.5	57.9	13.2	65.7	13.5	61.8	13.4	62.9	10.9	61.7	11.1	61.1	11.1	57.7	13.2	63.4	11.1	62.4	11.3	59.8
14	**G2**	10.4	62.8	10.6	63.5	11.0	58.7	11.6	56.7	11.2	60.7	11.7	57.2	14.5	63.8	14.2	61.3	15.2	60.0	11.0	58.7	11.2	60.0	11.1	57.5	13.0	61.3	11.1	57.8	11.3	58.7
15	**H1**	9.4	63.6	10.1	63.1	9.3	66.3	11.4	59.0	11.4	58.2	11.4	55.4	13.5	61.1	9.9	64.5	13.4	62.5	10.9	60.7	10.9	62.6	11.1	56.5	12.5	60.2	11.3	61.3	11.3	59.2
16	**H2**	9.0	64.3	10.9	64.6	10.2	62.2	11.3	58.3	11.1	58.0	11.5	57.8	14.1	61.3	12.3	61.3	14.6	60.8	11.0	57.2	11.2	58.3	10.9	63.2	12.7	59.8	11.2	58.3	11.2	61.7
17	**H3**	8.2	68.8	9.0	65.6	9.6	63.5	11.4	58.4	11.6	61.4	11.5	55.8	13.3	62.7	12.9	61.8	11.5	66.2	11.1	60.3	11.2	58.4	11.0	59.5	12.7	61.0	11.2	61.6	11.1	63.0
18	**H4**	9.9	68.0	13.7	61.7	11.9	62.9	10.9	62.1	11.5	58.6	11.3	59.1	13.8	63.3	13.9	60.2	16.8	59.8	11.0	56.5	11.2	61.9	11.1	58.9	12.7	60.7	11.1	57.1	11.2	59.4
19	**H5**	11.1	67.7	11.4	63.4	10.4	63.2	11.7	59.0	11.7	58.3	11.7	56.6	13.9	60.9	12.9	64.7	15.9	59.3	11.0	60.4	11.0	59.1	11.1	60.9	13.8	61.1	11.1	60.7	11.1	60.6
20	**H6**	12.0	63.9	10.8	66.5	9.6	63.8	11.9	56.2	11.6	58.3	11.7	58.1	12.8	63.2	15.3	60.4	15.2	62.1	11.1	60.5	11.2	61.7	11.1	59.2	13.9	60.6	11.1	64.1	11.5	60.8

4.7. AMYLOSE CONTENT DETERMINATION, THE 2006 SEASON

Amylose-in-starch content (amylose/amylopectin ratio, %) and respective total starch data analysis results are presented in Table 4.7.1. Results of analysis of high-amylose (70%) maize starch reference for each batch is also included. Only a few lines were analysed in the 2007 season because of a problem with a new batch of the Megazyme® amylose/amylopectin assay kit. The acquired data were not used in further data analysis.

The results of analysis are in agreement with expected *ca*. 20–25% amylose-in-starch content for wild-type starches (FAO, 1998; POWER, 2003), with lines DB and YA having slightly lower amylose content of 13%. Thus, none of the analysed lines had waxy or high-amylose starch phenotype. Consequently, successful selection for this trait at phenotypic level does not seem possible within the given set of triticale lines.

4.8. COMBINED ANALYSES OF THE 2006 SEASON DATA

Descriptive statistics and normality distribution tests for the traits of the 2006 season trials are presented in Tables 4.8.1 and 4.8.2. High coefficient of variation values for grain yield (CV = 29.66%) and starch yield (CV = 29.97%) are explained by the fact that the set of lines and cultivars under investigation was of different genetic background and bred for different purpose of use – green fodder, grazing, hay, grain for bread making, and dual purpose. Additionally, the Table 4.8.1 summarises data from locations of the two distinct environments, namely Swartland and Overberg, where precipitation conditions and rust disease pattern was different during the season (see Tables 4.1.1 and 4.4.16).

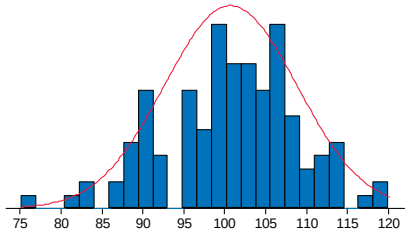
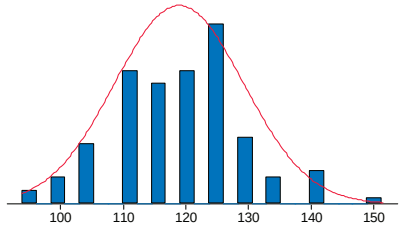
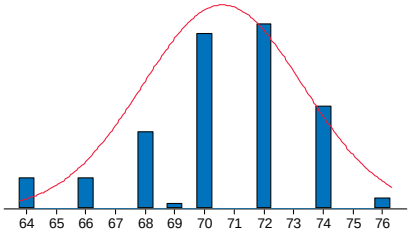
Table 4.7.1. Total starch and amylose-in-starch content for selected lines of the 2006 and 2007 Mariendahl elite trial

Location (-rep.)	Year	Entry No.	Name	Starch, % DWB	Amylose-in-starch, %	Batch reference: high amylose maize starch (70%)
ME	2006	1	**CA**	59.88	19.57	68.09
ME	2006	2	**CB**	55.71	17.39	67.84
ME	2006	3	**CC**	58.53	19.81	68.09
ME	2006	4	**CD**	61.61	20.18	68.09
ME	2006	5	**CE**	62.96	19.00	66.12
ME	2006	6	**D1**	64.10	20.24	67.84
ME	2006	7	**DB**	63.62	12.85	66.12
ME	2006	8	**YA**	63.18	13.10	66.12
ME	2006	9	**DC**	60.27	18.98	66.12
ME	2006	10	**YB**	62.65	18.51	66.12
ME	2006	11	**YC**	60.70	19.20	68.16
ME	2006	12	**DD**	62.85	21.06	68.16
ME	2006	13	**DE**	60.74	19.77	68.16
ME	2006	14	**DF**	62.54	20.94	68.16
ME	2006	15	**DG**	61.17	18.12	68.16
ME	2006	16	**DH**	59.95	19.68	68.16
ME	2006	17	**Y1**	60.55	20.77	68.16
ME	2006	18	**Y2**	61.05	20.22	68.16
ME	2006	19	**YD**	62.02	20.02	68.16
ME	2006	20	**Y3**	62.52	20.48	68.16
ME-2	2007	1	**CA**	67.49	16.84	63.23
ME-2	2007	2	**CC**	65.56	16.00	63.23
ME-2	2007	3	**CE**	65.03	15.10	63.23
ME-2	2007	4	**CD**	63.14	17.15	63.23
ME-2	2007	5	**D1**	63.88	16.86	63.23
ME-2	2007	6	**D2**	65.73	19.97	63.23
ME-2	2007	7	**D3**	67.12	17.01	63.23
ME-2	2007	8	**D4**	65.31	15.44	63.23
ME-2	2007	9	**YC**	63.65	15.35	63.23

Table 4.8.1. Descriptive statistics of the 2006 season traits averaged per entry and location

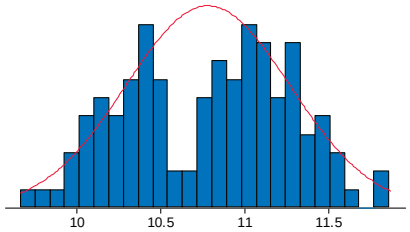
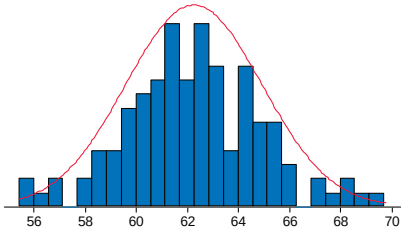
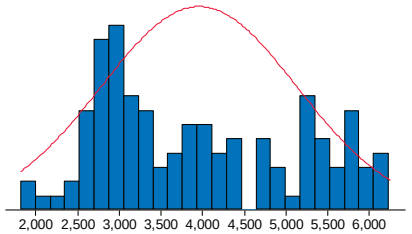
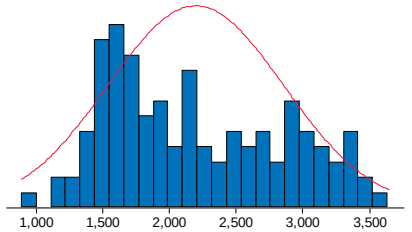
Statistic parameter	Days to heading	Height, cm	Test weight, kg.HL^{-1}	Moisture content, %	Starch content, %	Grain yield as-is, kg.ha^{-1}	Starch yield, kg.ha^{-1}
Mean	100.69	118.92	70.61	10.78	62.25	3954.04	2196.28
Median	101.14	119.85	70.44	10.76	62.17	3747.87	2085.69
Count	120	120	120	120	120	120	120
Range	43	55	12	2.1	13.69	4235.18	2630.89
Minimum	76	95	64	9.71	55.71	1903.67	945.89
Maximum	119	150	76	11.81	69.4	6138.85	3576.78
95% Confidence Interval for the Mean, from	99.23	117.05	70.12	10.69	61.77	3742.02	2077.28
95% Confidence Interval for the Mean, to	102.15	120.78	71.1	10.87	62.73	4166.07	2315.27
Standard Error	0.74	0.94	0.25	0.04	0.24	107.08	60.10
Standard Deviation	8.095	10.314	2.695	0.487	2.663	1172.964	658.315
Variance	65.526	106.380	7.265	0.237	7.090	1375845.29	433378.39
Coefficient of Variation, %	8.04	8.67	3.82	4.52	4.28	29.66	29.97
Kurtosis	0.149	0.056	0.159	–0.939	0.211	–1.166	–1.053
Skewness	–0.305	0.150	–0.613	–0.103	0.104	0.361	0.378

Table 4.8.2. Normality distribution tests of the traits of the 2006 season trials

Test name	Test statistics	p-level	Conclusion: (5%)	Distribution histogram
Days to heading				
Lilliefor's Test	0.051		No evidence against normality	
Shapiro-Wilkes W	0.9866	0.2846	Accept Normality	
D'Agostino Skewness	1.3956	0.1628	Accept Normality	
D'Agostino Kurtosis	0.5135	0.6076	Accept Normality	
D'Agostino Omnibus	2.2115	0.3310	Accept Normality	
Height				
Lilliefor's Test	0.111		Strong evidence against normality	
Shapiro-Wilkes W	0.9726	0.0151	Reject Normality	
D'Agostino Skewness	0.6969	0.4859	Accept Normality	
D'Agostino Kurtosis	0.3079	0.7581	Accept Normality	
D'Agostino Omnibus	0.5804	0.7481	Accept Normality	
Test weight				
Lilliefor's Test	0.119		Strong evidence against normality	
Shapiro-Wilkes W	0.9205	2.56E-06	Reject Normality	
D'Agostino Skewness	2.6741	0.0075	Reject Normality	
D'Agostino Kurtosis	0.5330	0.5940	Accept Normality	
D'Agostino Omnibus	7.4351	0.0243	Reject Normality	

(continued)

Table 4.8.2. (continued)

Test name	Test statistics	p-level	Conclusion: (5%)	Distribution histogram
Moisture content				
Lilliefor's Test	0.085		Sufficient evidence against normality	
Shapiro-Wilkes W	0.9728	0.0156	Reject Normality	
D'Agostino Skewness	0.4765	0.6337	Accept Normality	
D'Agostino Kurtosis	–3.7775	0.0002	Reject Normality	
D'Agostino Omnibus	14.4966	0.0007	Reject Normality	
Starch content				
Lilliefor's Test	0.053		No evidence against normality	
Shapiro-Wilkes W	0.9919	0.7083	Accept Normality	
D'Agostino Skewness	0.4821	0.6297	Accept Normality	
D'Agostino Kurtosis	0.6425	0.5206	Accept Normality	
D'Agostino Omnibus	0.6452	0.7243	Accept Normality	
Grain yield 'as-is'				
Lilliefor's Test	0.132		Strong evidence against normality	
Shapiro-Wilkes W	0.9290	8.29E-06	Reject Normality	
D'Agostino Skewness	1.6387	0.1013	Accept Normality	
D'Agostino Kurtosis	–6.1432	8.09E-10	Reject Normality	
D'Agostino Omnibus	40.4241	1.67E-09	Reject Normality	
Starch yield				
Lilliefor's Test	0.113		Strong evidence against normality	
Shapiro-Wilkes W	0.9423	6.12E-05	Reject Normality	
D'Agostino Skewness	1.7133	0.0867	Accept Normality	
D'Agostino Kurtosis	–4.7936	1.64E-06	Reject Normality	
D'Agostino Omnibus	25.9140	2.36E-06	Reject Normality	

Among the analysed traits only two (days to heading and starch content) were normally distributed, thus all subsequent data analyses were done using nonparametric methods (i.e. methods which do not assume or require normally distributed data). Traits of lines were assessed by means of Spearman' rank correlation in order to discover traits associations, especially those that are coupled with starch yield. Results of the correlation analysis for the 2006 season elite trials are presented in Table 4.8.3.

Table 4.8.3. Spearman' rank correlation (SRC) of the measured traits in the 2006 season

Trait	Statistic parameter	Days to heading	Height, cm	Test weight, kg.HL^{-1}	Moisture content, %	Starch content, %	Grain yield as-is, kg.ha^{-1}
Height, cm	SRC	**–0.093**	1				
	t-test value	–1.017					
	Significance level	N.S.					
	Probability > t	3.09E-01					
Test weight, kg.HL^{-1}	SRC	**0.117**	**–0.006**	1			
	t-test value	1.271	–0.061				
	Significance level	N.S.	N.S.				
	Probability > t	2.04E-01	9.52E-01				
Moisture content, %	SRC	**0.447**	**0.017**	**–0.137**	1		
	t-test value	4.878	0.188	–1.496			
	Significance level	***	N.S.	N.S.			
	Probability > t	1.07E-06	8.50E-01	1.35E-01			
Starch content, %	SRC	**–0.157**	**0.251**	**0.005**	**0.047**	1	
	t-test value	–1.717	2.736	0.053	0.517		
	Significance level	N.S.	**	N.S.	N.S.		
	Probability > t	8.59E-02	6.21E-03	9.58E-01	6.05E-01		
Grain yield as-is, kg.ha^{-1}	SRC	**0.556**	**0.140**	**0.165**	**0.680**	**0.146**	1
	t-test value	6.066	1.527	1.796	7.415	1.592	
	Significance level	***	N.S.	N.S.	***	N.S.	
	Probability > t	1.31E-09	1.27E-01	7.25E-02	1.22E-13	1.11E-01	
Starch yield, kg.ha^{-1}	SRC	**0.533**	**0.157**	**0.149**	**0.673**	**0.274**	**0.988**
	t-test value	5.813	1.716	1.626	7.336	2.994	10.778
	Significance level	***	N.S.	N.S.	***	**	***
	Probability > t	6.15E-09	8.61E-02	1.04E-01	2.20E-13	2.75E-03	4.38E-27

Remarks:

SRC values are corrected for ties.

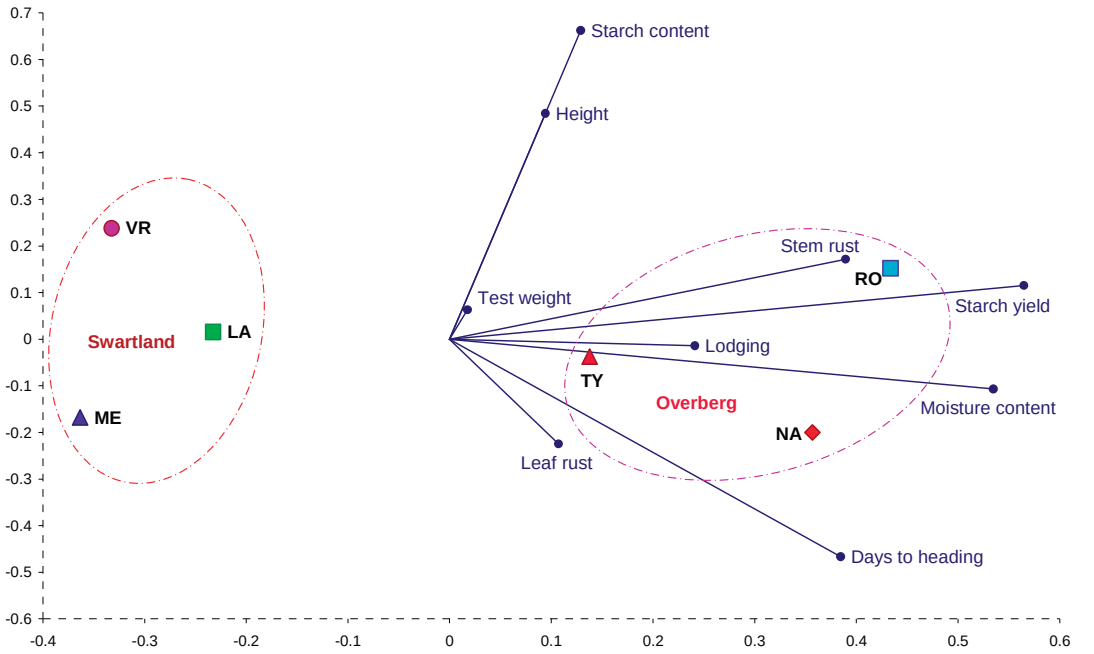
t-test values are for hypothesis r = 0.

Significance level (two-sided):
N.S. Not significant, P >0.05
** Significant, P <= 0.01
*** Significant, P <= 0.001

Starch yield was highly positively correlated with grain yield ($R^2 = 0.988$, $P < 0.001$). Both starch yield and grain yield were positively correlated with moisture content ($R^2 = 0.673$ and $R^2 = 0.680$, respectively; $P < 0.001$) and days to heading ($R^2 = 0.533$ and $R^2 = 0.556$, respectively; $P < 0.001$). Positive correlation with moisture content can be explained by some moisture being not accounted for by the method of moisture determination, which resulted in proportionally higher values of grain yield and starch yield. The other possible reason might be that the grain that was harvested from lines/locations with higher grain yield (late maturing) was exposed to later rains, thus the grain was not naturally dried to the same level as the other lower-yielding (earlier maturing) lines/locations which were not exposed to later rains. It seems that for the same reason days to heading are positively correlated with moisture content in grain. There is a positive correlation between yield and days to heading. The later-maturing lines benefit from the later rains and subsequently produce higher grain yield. Longer days to heading could be used as an indirect selection trait for higher grain and starch yields in these regions. However, this suggestion has to be used cautiously considering that cultivars with shorter days to heading, such as CD, are nevertheless characterised by relatively high yields in some locations. The unstable performance of the CD cultivar in some locations or years can be attributed to its susceptibility to rusts.

Principal component analysis of the traits and locations (Figure 4.8.1) distinguished two sub-environments – Swartland and Overberg. This was expected from prior observations. It suggests that breeding and deployment of distinct triticale cultivar types for these two sub-environments might be recommended for a better exploitation of the environmental and genotypic differences (genotype-by-environment interactions).

Figure 4.8.1. Principal component analysis of traits and locations of the 2006 season trials



4.8.1. Starch yield analysis, the 2006 season

The analysis of variance and breeding indices for starch yield are shown in Tables 4.8.1.1 and 4.8.1.2.

Table 4.8.1.1. ANOVA of starch yield of the 2006 season trials

Source	DF	SS	MS	F	P
Entry	19	1855354	97650.24	1.460792	0.11809
Location	5	43366166	8673233	129.7467	0.00001
Interaction (Error)	95	6350507	66847.44		

Other statistic parameters:

Locations	6	**LSD(5%)**	279.57
Mean	2196.278	**Pairwise SE**	149.273
Std Error (SE)	258.549	**CV%**	11.77
Std Deviation (SD)	127.575	**Mean/LSD**	7.856
Vp	16275.38	**H^2=Vg/Vp**	0.315
Vg	5134.139	**G/GGE% (SS)**	22.61
MSe	66847.45	**G/GGE% (VC)**	7.13

Table 4.8.1.2. Starch yield ranks and breeding indices across locations of the 2006 elite trials

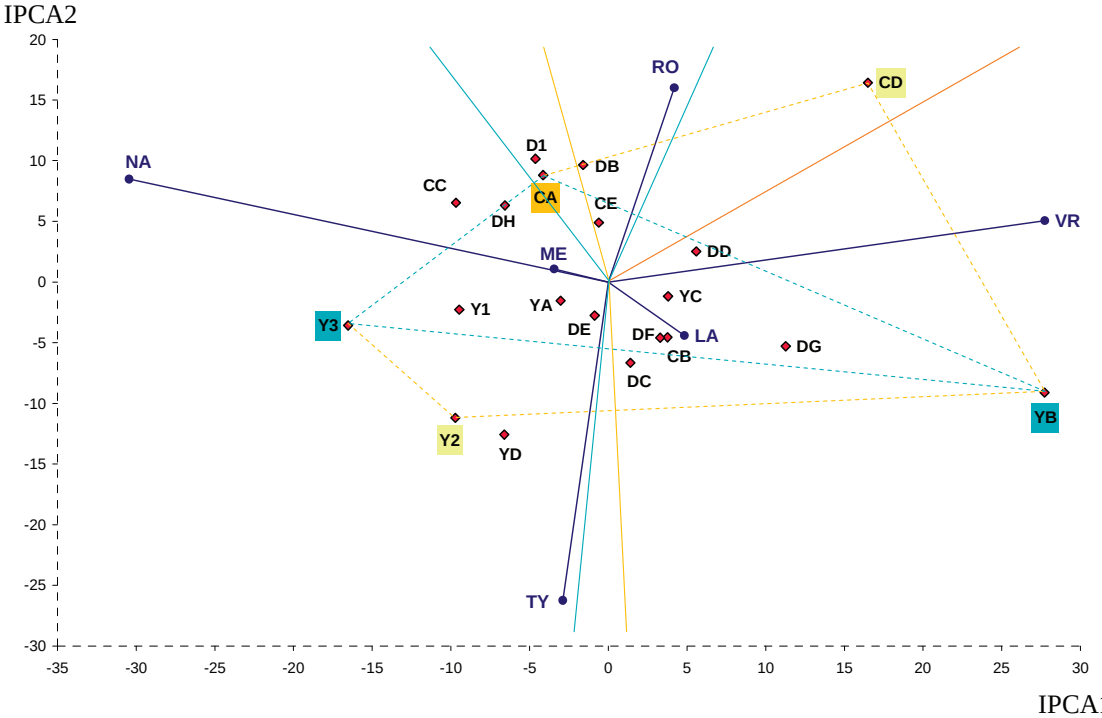
Entry	Mean, kg.ha^{-1}	RV, %	HARV, %	SI, %	HASI, %	Mean/ LSD5%	Mean/ LSD1%
CA	2395.48	109	103	100	100	8.57	6.33
Y3	2379.03	108	103	99	100	8.51	6.28
YC	2335.96	106	102	98	99	8.36	6.17
CC	2288.91	104	101	96	99	8.19	6.05
Y1	2274.83	104	101	95	98	8.14	6.01
YA	2274.01	104	101	95	98	8.13	6.01
YB	2263.77	103	101	95	98	8.10	5.98
Y2	2260.43	103	101	94	98	8.09	5.97
DC	2236.51	102	101	93	98	8.00	5.91
DB	2200.61	100	100	92	97	7.87	5.81
YD	2199.99	100	100	92	97	7.87	5.81
DE	2183.52	99	100	91	97	7.81	5.77
CE	2173.86	99	100	91	97	7.78	5.74
DH	2149.09	98	99	90	97	7.69	5.68
DF	2136.85	97	99	89	97	7.64	5.64
DD	2125.56	97	99	89	97	7.60	5.61
D1	2123.98	97	99	89	97	7.60	5.61
CD	2088.81	95	98	87	96	7.47	5.52
DG	1944.96	89	97	81	94	6.96	5.14
CB	1889.42	86	96	79	93	6.76	4.99

Remarks:
Entries are significantly different at the 5% or 1% level if their Mean/LSD5% or Mean/LSD1% differ by 1.0 or more.
RV = Relative Value; HARV = Heritability Adjusted Relative Value.
SI = Superior Index or Value Relative to Maximum with 100 indicating the best.
HASI = Heritability Adjusted Superior Index.
HARV and HASI are recommended when evaluating varieties across environments.

Top-17 lines for starch yield were not statistically different at the 5% level of significance across locations, with the mean starch yield ranging between 2124–2395kg.ha^{-1} for these lines. Broad-sense heritability (H^2) for starch yield was 0.315. The results of the 2006 season cross-site analysis of starch yield for genotype-by-environment (G×E) interactions using an additive main effects and multiplicative interaction (AMMI) model are presented in Addendum 6 and are summarised together by principal components biplot of AMMI2 model (Figure 4.8.1.1). Additional information in the form of colouring is added into the biplot for easier interpretability. The same approach was used for the interpretation of other biplots in the following sections. Basic guidelines for a G×E principal components interaction biplot

interpretation see in Table 3.11.1. The Figure 4.8.1.1 shows that lines YB, Y3 and CD are characterised by a higher positive G×E interaction (high specific adaptation, or narrowly adapted) for the starch yield in certain locations (Langgewens and Vredenburg for YB; Roodebloem and Vredenburg for CD; Tygerhoek, Napier and Mariendahl for Y3) compared to lines DE, CE and YC (situated closer to the center of the biplot) which demonstrated higher levels of stability for starch yield across locations at the expense of its lower average yield. The lines DE, CE and YC were therefore characterised by a broader (or wide) adaptation. The line CA was characterised by high average starch yields coupled with relative stability across trials located at the border of the two sub-environments. It has to be reminded that stability of any sort depends on the locations and the genotypes included in the particular experiment. A genotype that is stable in one set of environments may not be stable in another; similarly, a stable genotype may not be stable if evaluated with a different set of genotypes (ANNICCHIARICO, 2002).

Figure 4.8.1.1. Two-way interaction biplot of the AMMI2 model for the 2006 season starch yield



The best genotypes for each location suggested by the AMMI2 model are presented in Table 4.8.1.3 and Figure 4.8.1.2. For the Swartland region lines YB and CA could be recommended (Y3 is not recommended because it performs well only in location Mariendahl, which is not representative for the Swartland subenvironment as a whole), and for the Overberg lines Y3, CA and Y2. However, situation with the Overberg region was not clear because none of these lines gave consistently highest starch yield across at least two locations of the region. It might be suggested that combination by breeding of the yield potential and its stability traits of these lines could produce lines which are better adapted for the target regions.

Table 4.8.1.3. The best genotypes of the 2006 season for starch yield suggested by the AMMI2 model with high yield potential and adaptation

(values fitted for the G×E interaction, kg.ha^{-1})

Location / Score	Swartland			Overberg		
	VR	LA	ME	RO	TY	NA
1st	**YB** 2532	**YB** 1950	**Y3** 1721	**CA** 3471	**Y2** 2640	**Y3** 3410
2nd	-	-	**CA** 1709	**CD** 3363	**Y3** 2587	-
3rd	**CD** 2184	**CA** 1859	-	-	-	-

Figure 4.8.1.2. Comparison of the best four starch yielding triticale lines suggested by the AMMI2 model for the 2006 season across locations

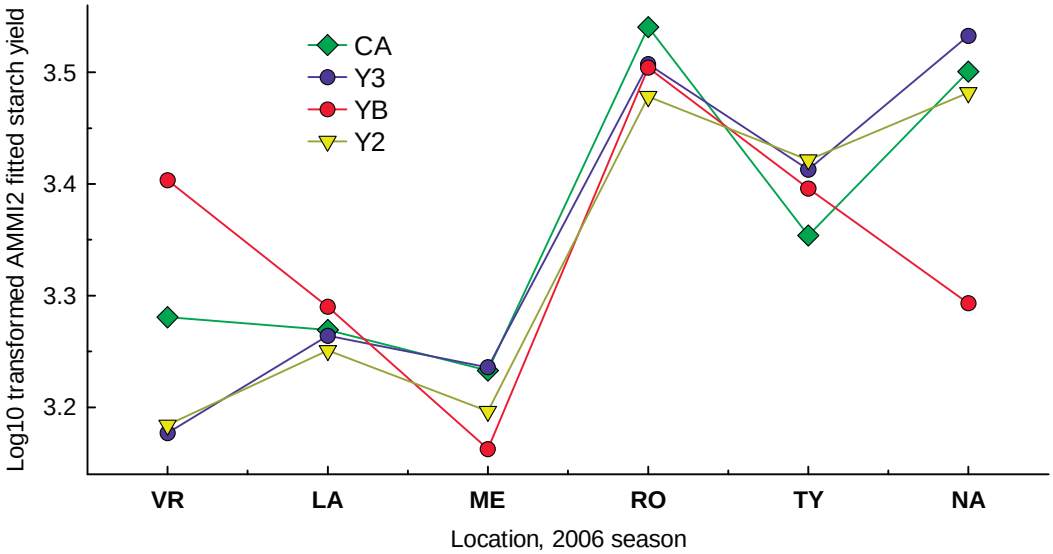
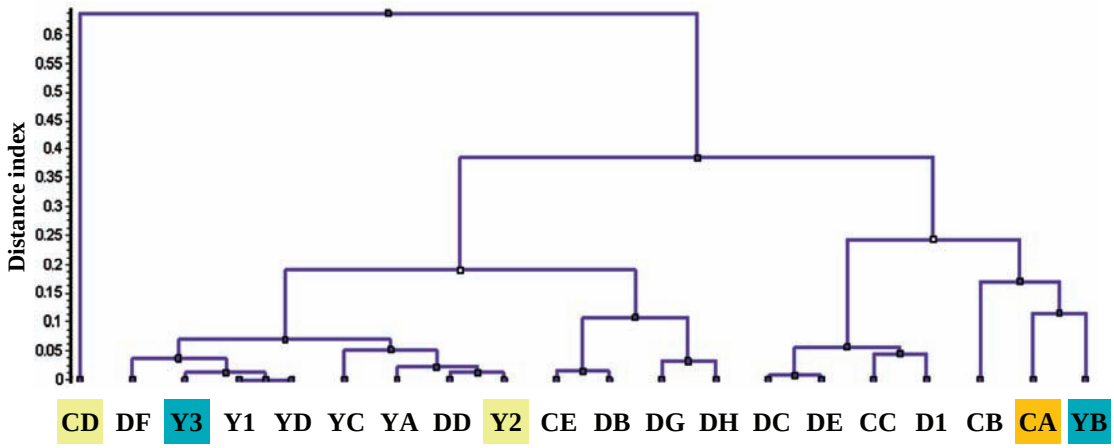


Figure 4.8.1.3 shows the hierarchical agglomerative clustering (HAC) dendrogram of the triticale lines. The dendrogram was developed from the traits described in Table 4.8.3. The clystering dendrogram can be used for the selection of lines for the crossing block. It can be seen that lines CA and YB show similarities in their traits expression and both these lines are characterised by a high starch yield in the Swartland region. The combination of their genetic material by breeding seems to be promising for the development of a cultivar more adapted for the region environment. On the other hand, inter-crossing of lines Y3, Y2 and CA could result in high-starch-yielding lines better adapted for the Overberg region. Considering results of the 2006 season starch yield, lines YB and CA could be recommended as an initial material for the development of new high-starch-yielding cultivars for the Swartland region, and lines Y3, CA and Y2 for the Overberg region.

Figure 4.8.1.3. Hierarchical agglomerative clustering (HAC) dendrogram of triticale lines for the 2006 season



4.9. COMBINED ANALYSIS OF THE 2007 SEASON DATA

Descriptive statistics and normality distribution tests for the 2007 season traits are presented in Tables 4.9.1 and 4.9.2. Only two traits (plants height and grain moisture content) were normally distributed. The 2007 season was characterised by a

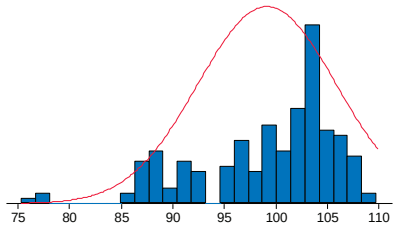
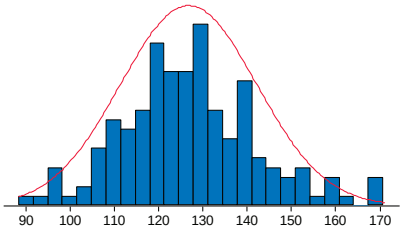
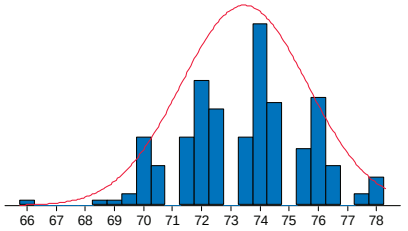
generally higher starch yield (2952–3142kg.ha^{-1}, 95%CI; Table 4.9.1) compared to the 2006 season (2077–2315kg.ha^{-1}, 95%CI; Table 4.8.1). Correlation analysis of the 2007 season traits is presented in Table 4.9.3. Correlation of starch yield with other traits was in agreement with the 2006 season results, however correlation between starch yield and test weight in the 2007 season was significantly higher ($R^2 = 0.324$, $P < 0.001$). Starch yield was strongly positively correlated with grain yield ($R^2 = 0.975$, $P < 0.001$) and had a weak positive correlation with days to heading ($R^2 = 0.400$, $P < 0.001$) and moisture content ($R^2 = 0.420$, $P < 0.001$).

In the 2007 season, it was possible to assess traits that are directly related to the research question, namely ethanol output and ethanol yield. Correlation pattern of the ethanol yield with other traits closely follows the correlation pattern for the starch yield, both traits being highly positively correlated with each other ($R^2 = 0.910$, $P < 0.001$). Dispite of this, ethanol yield did not show any significant correlation with total starch content, as was expected (JACOBI & HARTMANN, 2005). On the other hand, both starch yield and ethanol yield had a weak positive correlation with protein content, which was completely opposite to the expected from data of other research studies (RIFFKIN *et al.*, 1990; SWANSTON *et al.*, 2007; KINDRED *et al.*, 2008; DAVIS-KNIGHT & WEIGHTMAN, 2008). Ethanol output had shown a weak but significant positive correlation with total starch content ($R^2 = 0.359$, $P < 0.001$) and moisture content ($R^2 = 0.461$, $P < 0.001$), moderate correlation with ethanol yield ($R^2 = 0.568$, $P < 0.001$), being negatively correlated with protein content ($R^2 = –0.154$, $P < 0.05$), days to heading ($R^2 = –0.316$, $P < 0.001$) and plant height ($R^2 = –0.448$, $P < 0.001$).

Table 4.9.1. Descriptive statistics for the 2007 season traits averaged per entry and location

Statistic parameter	Days to heading	Height, cm	Test weight, kg.HL^{-1}	PSI, %	Protein content, %	Moisture content, %	Starch content DWB, %	Grain yield as-is, kg.ha^{-1}	Starch yield DWB, kg.ha^{-1}	Ethanol output as-is, L.tonne^{-1}	Ethanol yield as-is, L.ha^{-1}
Mean	99.12	126.76	73.46	64.47	10.61	10.85	63.90	5352.49	3047.31	471.63	2535.71
Median	101.50	126.00	74.00	64.37	11.04	10.87	64.16	5502.78	3121.25	472.03	2563.98
Count	180	160	180	180	180	180	180	180	180	180	180
Range	33	79	12	18.59	9.97	1.99	9.36	7275.85	4239.68	175.41	3645.44
Minimum	76	90	66	56.93	5.07	10.03	58.51	648.97	362.70	402.60	275.12
Maximum	109	169	78	75.52	15.05	12.02	67.87	7924.82	4602.38	578.01	3920.56
95% Confidence Interval for the Mean, from	98.14	124.35	73.14	63.85	10.29	10.79	63.57	5186.02	2952.24	465.88	2446.30
95% Confidence Interval for the Mean, to	100.11	129.16	73.78	65.09	10.94	10.91	64.23	5518.95	3142.38	477.38	2625.13
Standard Error	0.50	1.22	0.16	0.31	0.17	0.03	0.17	84.36	48.18	2.91	45.31
Standard Deviation	6.692	15.420	2.168	4.206	2.231	0.397	2.243	1131.81	646.36	39.08	607.94
Variance	44.778	237.770	4.699	17.686	4.977	0.158	5.031	1280994.2	417777.6	1526.936	369590.47
Coefficient of Variation, %	6.75	12.16	2.95	6.52	21.02	3.66	3.51	21.15	21.21	8.29	23.98
Kurtosis	0.524	0.272	–0.077	–0.686	–0.261	0.043	–0.577	1.355	1.557	–0.301	0.738
Skewness	–1.024	0.333	–0.206	0.299	–0.563	0.122	–0.423	–0.789	–0.713	0.388	–0.519

Table 4.9.2. Normality distribution tests for the traits of the 2007 season

Test name	Test Statistics	p-level	Conclusion: (5%)	Distribution histogram
Days to heading				
Lilliefor's Test	0.110		Strong evidence against normality	
Shapiro-Wilkes W	0.9005	1.24E-09	Reject Normality	
D'Agostino Skewness	4.9158	8.84E-07	Reject Normality	
D'Agostino Kurtosis	1.3869	0.1655	Accept Normality	
D'Agostino Omnibus	26.0889	2.16E-06	Reject Normality	
Height				
Lilliefor's Test	0.067		Suggestive evidence against normality	
Shapiro-Wilkes W	0.9867	0.1333	Accept Normality	
D'Agostino Skewness	1.7381	0.0822	Accept Normality	
D'Agostino Kurtosis	0.8262	0.4087	Accept Normality	
D'Agostino Omnibus	3.7036	0.1570	Accept Normality	
Test weight				
Lilliefor's Test	0.072		Sufficient evidence against normality	
Shapiro-Wilkes W	0.9802	0.0117	Reject Normality	
D'Agostino Skewness	1.1487	0.2507	Accept Normality	
D'Agostino Kurtosis	–0.0735	0.9414	Accept Normality	
D'Agostino Omnibus	1.3249	0.5156	Accept Normality	

(continued)

Table 4.9.2. (continued)

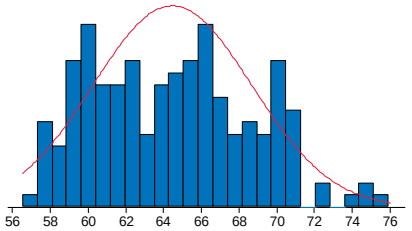
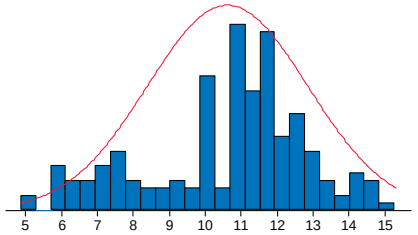
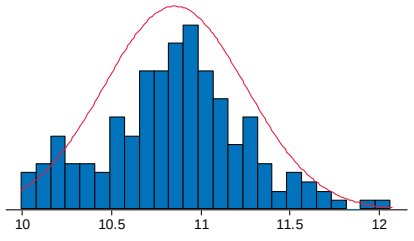
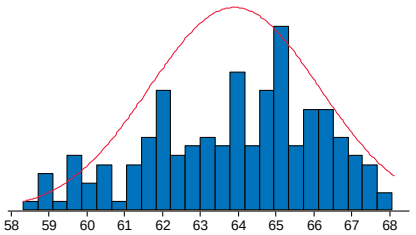
Test name	Test Statistics	p-level	Conclusion: (5%)	Distribution histogram
PSI				
Lilliefor's Test	0.068		Sufficient evidence against normality	
Shapiro-Wilkes W	0.9728	0.0014	Reject Normality	
D'Agostino Skewness	1.6553	0.0979	Accept Normality	
D'Agostino Kurtosis	–2.7634	0.0057	Reject Normality	
D'Agostino Omnibus	10.3764	0.0056	Reject Normality	
Protein content				
Lilliefor's Test	0.135		Strong evidence against normality	
Shapiro-Wilkes W	0.9516	8.01E-06	Reject Normality	
D'Agostino Skewness	2.9892	0.0028	Reject Normality	
D'Agostino Kurtosis	–0.6983	0.4850	Accept Normality	
D'Agostino Omnibus	9.4230	0.0090	Reject Normality	
Moisture content				
Lilliefor's Test	0.040		No evidence against normality	
Shapiro-Wilkes W	0.9875	0.1101	Accept Normality	
D'Agostino Skewness	0.6832	0.4945	Accept Normality	
D'Agostino Kurtosis	0.2772	0.7816	Accept Normality	
D'Agostino Omnibus	0.5436	0.7620	Accept Normality	
Starch content				
Lilliefor's Test	0.082		Strong evidence against normality	
Shapiro-Wilkes W	0.9693	0.0005	Reject Normality	
D'Agostino Skewness	2.3028	0.0213	Reject Normality	
D'Agostino Kurtosis	–2.1138	0.0345	Reject Normality	
D'Agostino Omnibus	9.7710	0.0076	Reject Normality	

Table 4.9.2. (continued)

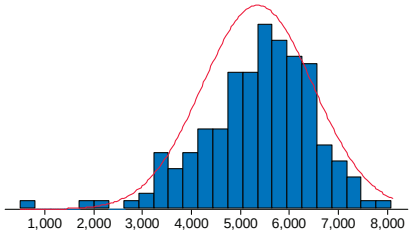
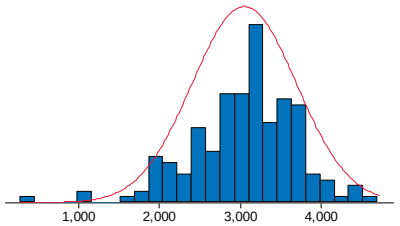
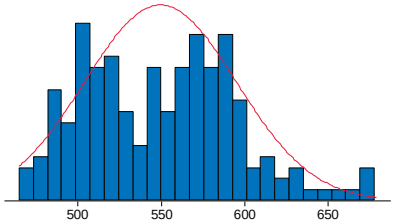
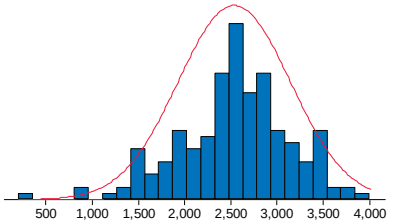
Test name	Test Statistics	p-level	Conclusion: (5%)	Distribution histogram
Grain yield				
Lilliefor's Test	0.072		Sufficient evidence against normality	
Shapiro-Wilkes W	0.9678	0.0004	Reject Normality	
D'Agostino Skewness	3.9963	6.43E-05	Reject Normality	
D'Agostino Kurtosis	2.6617	0.0078	Reject Normality	
D'Agostino Omnibus	23.0554	9.85E-06	Reject Normality	
Starch yield				
Lilliefor's Test	0.079		Strong evidence against normality	
Shapiro-Wilkes W	0.9692	0.0005	Reject Normality	
D'Agostino Skewness	3.6727	0.0002	Reject Normality	
D'Agostino Kurtosis	2.8979	0.0038	Reject Normality	
D'Agostino Omnibus	21.8867	1.77E-05	Reject Normality	
Ethanol output				
Lilliefor's Test	0.084		Strong evidence against normality	
Shapiro-Wilkes W	0.9695	0.0006	Reject Normality	
D'Agostino Skewness	2.1237	0.0337	Reject Normality	
D'Agostino Kurtosis	–0.8497	0.3955	Accept Normality	
D'Agostino Omnibus	5.2320	0.0731	Accept Normality	
Ethanol yield				
Lilliefor's Test	0.076		Strong evidence against normality	
Shapiro-Wilkes W	0.9804	0.0125	Reject Normality	
D'Agostino Skewness	2.7781	0.0055	Reject Normality	
D'Agostino Kurtosis	1.7753	0.0758	Accept Normality	
D'Agostino Omnibus	10.8698	0.0044	Reject Normality	

Table 4.9.3. Spearman' rank correlation (SRC) of the measured traits in the 2007 season

Trait	Statistic parameter	Days to heading	Height, cm	Test weight, kg.HL^{-1}	PSI, %	Protein content, %	Moisture content, %	Starch content, %	Grain yield as-is, kg.ha^{-1}	Starch yield, kg.ha^{-1}	Ethanol output as-is, L.tonne^{-1}
Height, cm	SRC	**0.370**	1								
	t-test value	4.663									
	Significance level	***									
	Probability > t	3.11E-06									
Test weight, kg.HL^{-1}	SRC	**0.170**	**0.085**	1							
	t-test value	2.269	1.075								
	Significance level	*	N.S.								
	Probability > t	2.33E-02	2.83E-01								
PSI, %	SRC	**–0.526**	**–0.409**	**–0.116**	1						
	t-test value	–7.037	–5.162	–1.558							
	Significance level	***	***	N.S.							
	Probability > t	1.97E-12	2.45E-07	1.19E-01							
Protein content, %	SRC	**0.197**	**0.135**	**0.140**	**–0.676**	1					
	t-test value	2.638	1.699	1.877	–9.040						
	Significance level	**	N.S.	N.S.	***						
	Probability > t	8.35E-03	8.93E-02	6.05E-02	1.57E-19						
Moisture content, %	SRC	**–0.028**	**–0.234**	**0.087**	**–0.260**	**0.414**	1				
	t-test value	–0.376	–2.948	1.170	–3.481	5.540					
	Significance level	N.S.	**	N.S.	***	***					
	Probability > t	7.07E-01	3.20E-03	2.42E-01	5.00E-04	3.02E-08					
Starch content, %	SRC	**–0.365**	**–0.269**	**0.176**	**0.575**	**–0.360**	**–0.120**	1			
	t-test value	–4.887	–3.387	2.360	7.687	–4.811	–1.608				
	Significance level	***	***	*	***	***	N.S.				
	Probability > t	1.02E-06	7.06E-04	1.83E-02	1.50E-14	1.50E-06	1.08E-01				
Grain yield as-is, kg.ha^{-1}	SRC	**0.484**	**0.040**	**0.279**	**–0.414**	**0.286**	**0.453**	**–0.055**	1		
	t-test value	6.482	0.504	3.737	–5.539	3.824	6.062	–0.733			
	Significance level	***	N.S.	***	***	***	***	N.S.			
	Probability > t	9.07E-11	6.14E-01	1.86E-04	3.04E-08	1.31E-04	1.34E-09	4.64E-01			

(continued)

Table 4.9.3. (continued)

Trait	Statistic parameter	Days to heading	Height, cm	Test weight, kg.HL^{-1}	PSI, %	Protein content, %	Moisture content, %	Starch content, %	Grain yield as-is, kg.ha^{-1}	Starch yield, kg.ha^{-1}	Ethanol output as-is, L.tonne^{-1}
Starch yield, kg.ha^{-1}	SRC	**0.400**	**–0.031**	**0.324**	**–0.300**	**0.222**	**0.420**	**0.133**	**0.975**	1	
	t-test value	5.353	–0.390	4.330	–4.020	2.972	5.621	1.784	13.049		
	Significance level	***	N.S.	***	***	**	***	N.S.	***		
	Probability > t	8.66E-08	6.97E-01	1.49E-05	5.82E-05	2.96E-03	1.90E-08	7.44E-02	6.43E-39		
Ethanol output as-is, L.tonne^{-1}	SRC	**–0.316**	**–0.448**	**0.024**	**0.245**	**–0.154**	**0.461**	**0.359**	**0.181**	**0.248**	1
	t-test value	–4.231	–5.646	0.315	3.281	–2.059	6.165	4.805	2.428	3.322	
	Significance level	***	***	N.S.	**	*	***	***	*	***	
	Probability > t	2.33E-05	1.64E-08	7.53E-01	1.03E-03	3.95E-02	7.06E-10	1.55E-06	1.52E-02	8.94E-04	
Ethanol yield as-is, L.ha^{-1}	SRC	**0.261**	**–0.185**	**0.238**	**–0.227**	**0.149**	**0.554**	**0.108**	**0.895**	**0.910**	**0.568**
	t-test value	3.492	–2.327	3.190	–3.033	1.999	7.406	1.438	11.976	12.173	7.604
	Significance level	***	*	**	**	*	***	N.S.	***	***	***
	Probability > t	4.79E-04	2.00E-02	1.42E-03	2.42E-03	4.56E-02	1.31E-13	1.50E-01	4.75E-33	4.31E-34	2.87E-14

Remarks:

SRC values are corrected for ties.

t-test values are for hypothesis r = 0.

Significance level (two-sided):

N.S. Not significant, P >0.05
* Significant, P <= 0.05
** Significant, P <= 0.01
*** Significant, P <= 0.001

Positive correlation of total starch content with ethanol yield was demonstrated by other researches (JACOBI & HARTMANN, 2005). In some studies, triticale yielded less ethanol per unit of starch content and was less responsive to increases in starch content compared to wheat and rye (ROSENBERGER, 2005). Similarly, RIFFKIN *et al.* (1990) could not use starch content alone as an accurate predictor for ethanol yield in their research. Weak correlation of ethanol output with total starch content in our study could have two possible explanations: not all the available starch was converted into ethanol; other carbohydrates (e.g. NSP – the third major component of grain after starch and protein), which were not accounted for by the total starch assay, were converted into ethanol. The latter is especially possible because OPTIMASH™ VR cellulose and hemicellulose hydrolysing enzymes were added to the mash, thus aiding for the break down of fibrous particles leading to release of enclosed polysaccharides. It was argued that for dry-grind process high ethanol yield ('high total fermentable') is a more accurate indicator of grain quality than total starch content (ANONYMOUS, 2003; BOTHAST & SCHLICHER, 2005). The question is further discussed in section 4.10.1 "Ethanol output analysis."

It can be suggested that positive correlation of ethanol output with moisture content is explainable by more porous (softer or mealy) grain endosperm of some lines, considering positive correlation of the ethanol output with PSI, $R^2 = 0.245$, $P < 0.01$. These softer lines could possibly have higher content of friabilin (puroindolins) matrix proteins (GREENWELL & SCHOFIELD, 1989; MORRIS *et al.*, 1994; ODA & SCHOFIELD, 1997; BALDWIN, 2001). In such case protein matrix could be more loosely attached to the starch granules which provides air spaces within the endosperm contrary to the steely (vitreous) endosperm with a tightly packed matrix of protein, starch and cell walls (SMITH *et al.*, 2006). Because of this, starch granules

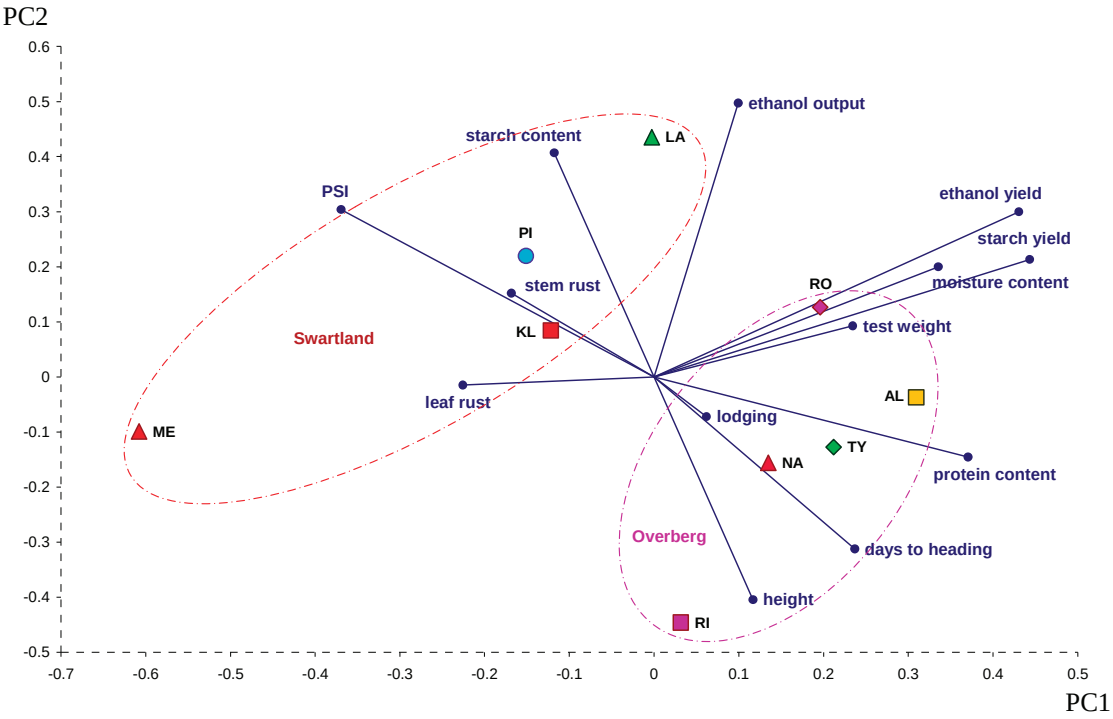
of softer grain lines could be characterised by a larger effective hydrophilic surface before and after milling, where molecules of water could bind and starch hydrolysing enzymes could get easier access, which in turn resulted in higher ethanol outputs. This suggestion is particularly reasonable considering that in our study the 'no-heat' fermentation method was used, which does not require complete gelatinisation of the starch granules.

Correlation values of total starch content ($R^2 = 0.108$, $P > 0.05$) and protein content ($R^2 = 0.149$, $P < 0.05$) with ethanol yield did not confirm the hypothesis that they are highly correlated, thus these traits cannot serve as reliable predictors for the ethanol yield. Relative moisture content had shown better correlations with starch yield ($R^2 = 0.42$, $P < 0.001$), ethanol output ($R^2 = 0.461$, $P < 0.001$) and ethanol yield ($R^2 = 0.554$, $P < 0.001$), thus could be cautiously considered as an indirect selection trait for these traits. It has to be noted that the relative moisture content has to be measured after a period of equalisation for few weeks in a store room. However, much better predictors for ethanol yield would be grain yield ($R^2 = 0.895$, $P < 0.001$) and starch yield ($R^2 = 0.91$, $P < 0.001$).

Principal components analysis of traits and locations of the 2007 season (Figure 4.9.1, developed from traits described in Table 4.9.1) confirmed that there are two sub-environments, namely Swartland and Overberg regions, which are characterised by a different traits expression pattern in triticale lines under investigation, thus distinct set of cultivars better adapted for these sub-environments is required. The PCA had also shown that Tygerhoek was overlapping with other locations of the Overberg region thus could be considered being redundant trial location in the 2007 season. On the other hand, in the Swartland region Mariendahl was distinctively different from the other locations, thus could not be recommended as

a representative breeding and testing environment for the target region as a whole. It could be recommended that the Tygerhoek and Mariendahl trials locations being relocated to another testing sites, or resources allocated for these trial locations being reallocated for another research activities.

Figure 4.9.1. PCA of traits and locations of the 2007 season trials



The analysis of variance and breeding indices for starch yield are shown in Tables 4.9.4 and 4.9.5.

Table 4.9.4. ANOVA table for the 2007 season starch yield across locations

Source	DF	SS	MS	F-ratio	P-value
Entry	19	13622545	716976.1	5.547378	0.00001
Location	8	41514264	5189283.0	40.150450	0.00001
Interaction (Error)	152	19645382	129245.9		

Other statistic parameters:

Locations	9	**LSD(5%)**	342.33
Mean	3047.31	**Pairwise SE**	169.474
Std Error (SE)	359.508	**CV%**	11.8
Std Deviation (SD)	282.251	**Mean/LSD**	8.902
Vp	79665.63	**H^2=Vg/Vp**	0.82
Vg	65304.97	**G/GGE% (SS)**	40.95
MSe	129245.9	**G/GGE% (VC)**	33.57

Table 4.9.5. Breeding indices for starch yield across locations of the 2007 elite trials

Entry	Mean	RV%	HARV%	SI%	HASI%	Mean/LSD5%	Mean/LSD1%
G2	3364.49	110	108	100	100	9.83	7.44
D2	3282.91	108	107	98	98	9.59	7.26
G1	3266.81	107	106	97	98	9.54	7.22
D4	3263.19	107	106	97	98	9.53	7.22
H3	3254.14	107	106	97	98	9.51	7.20
D1	3215.58	106	105	96	97	9.39	7.11
H6	3193.20	105	104	95	96	9.33	7.06
D3	3160.77	104	103	94	95	9.23	6.99
H2	3148.39	103	102	94	95	9.20	6.96
H1	3147.85	103	102	94	95	9.20	6.96
EA	3132.22	103	102	93	94	9.15	6.93
YC	3121.84	102	102	93	94	9.12	6.90
Y2	3066.25	101	101	91	93	8.96	6.78
CC	3036.34	100	100	90	92	8.87	6.71
CA	2977.11	98	98	88	90	8.70	6.58
H4	2953.56	97	98	88	90	8.63	6.53
CE	2926.89	96	97	87	89	8.55	6.47
EB	2657.40	87	89	79	83	7.76	5.88
H5	2583.50	85	88	77	81	7.55	5.71
CD	2193.77	72	77	65	71	6.41	4.85

Remarks:

Entries are significantly different at the 5% or 1% level if their Mean/LSD5% or Mean/LSD1% differ by 1.0 or more.

RV = Relative Value; HARV = Heritability Adjusted Relative Value.

SI = Superior Index or Value Relative to Maximum with 100 indicating the best.

HASI = Heritability Adjusted Superior Index.

HARV and HASI are recommended when evaluating varieties across environments.

Top-14 starch yielding lines of the 2007 season were not statistically different at the 5% level across locations, similarly to the 2006 season results. Mean starch yield ranged between 3036–3364kg.ha^{-1} for these 14 lines, broad-sense heritability $H^2 = 0.82$. The results of cross-site analysis of the 2007 season starch yield for the G×E interaction using the AMMI model are presented in Addendum 7 and are summarised by a principal components biplot of the AMMI2 model in Figure 4.9.2. The figure shows that lines G2, D3, CD and EA were characterised by a high positive G×E interaction for the starch yield in some trial locations (Roodebloem and Napier

for lines G2 and D3; Mariendahl for EA; Piketberg and Klipheuwel for cultivar CD), compared to lines D1 and D2 (located closer to the centre of the biplot) with a lesser G×E interaction, which were characterised by a higher stability but lower starch yield in those locations. The line G2 was characterised by a high starch yield coupled with its relatively high stability across locations, especially in the Overberg region. The best lines in terms of starch yield for each location are presented in Table 4.9.6. The best lines in terms of starch yield in the 2007 season for the Swartland region were H3, G2 and EA, and for the Overberg region G2 and D2. These lines can be recommended as an initial material for the breeding programme as donors of high starch yield.

Figure 4.9.2. Two-way interaction biplot of the AMMI2 model for starch yield, the 2007 season

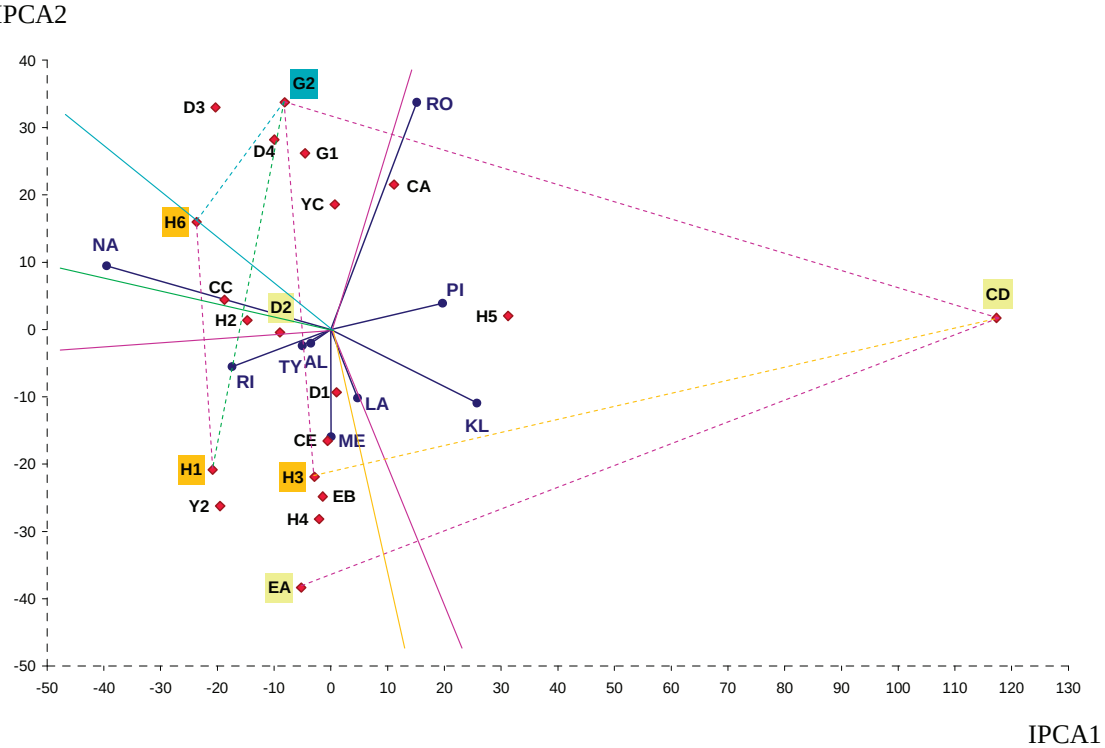


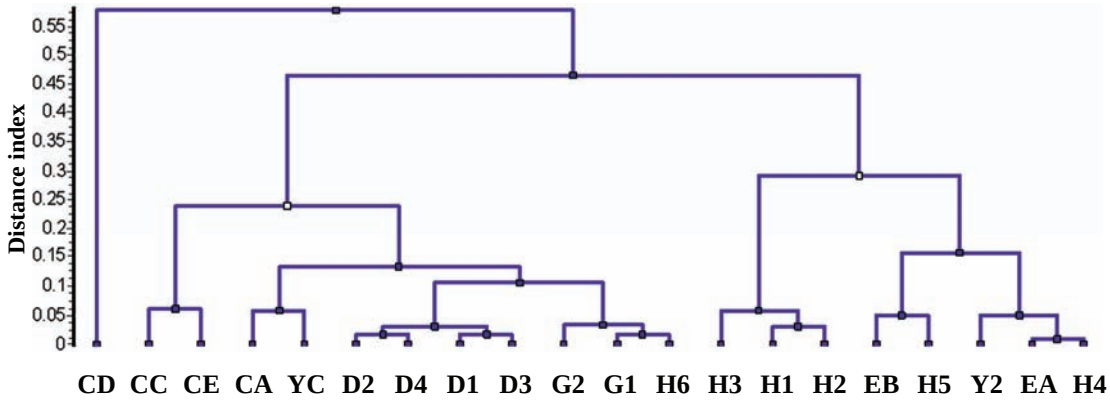
Table 4.9.6. The best genotypes of the 2007 season for starch yield suggested by the AMMI2 model with high yield potential and adaptation

(values fitted for G×E interaction, $kg.ha^{-1}$)

Location / Score	PI	KL	LA	ME	RO	TY	NA	RI	AL
1st	G2 3275	CD 3072	H3 3605	H3 2400	G2 4602	G2 3415	H6 3785	H1 3200	G2 3723
2nd	D2 3129	-	EA 3550	EA 2390	-	D2 3371	G2 3766	D2 3199	D2 3672
3rd	H3 3116	H3 2920	-	-	-	H3 3351	-	G2 3194	H3 3653

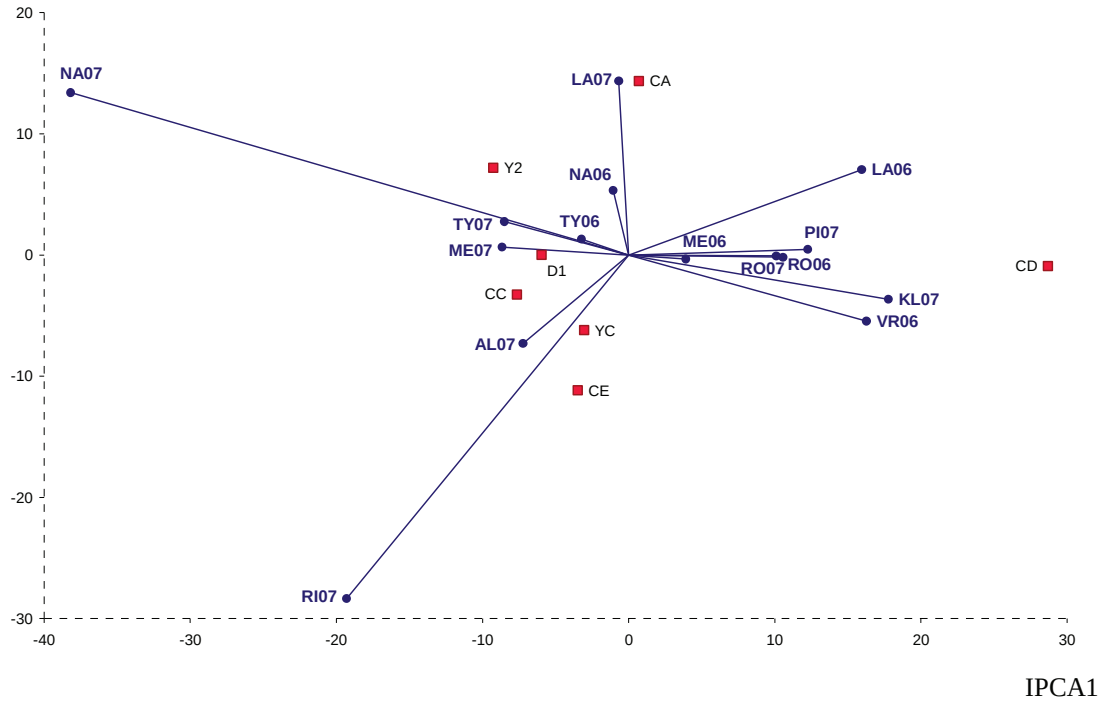
Considering Figure 4.9.3 (which was developed from traits described in Table 4.9.1), it could be expected that germplasm derived from breeding combinations H3 / EA and H3 / G2 could yield better adapted and high starch yielding lines for the Swartland region, as well as G2 / D2 for the Overberg region. It could be expected that G2 / H3 and D2 / H3 breeding combinations could result in breeding material better adapted for both sub-environments, taking into account distance index (Figure 4.9.3) of parents in these breeding combinations.

Figure 4.9.3. Hierarchical agglomerative clustering (HAC) dendrogram of triticale lines for the 2007 season



Combined analysis of the starch yield G×E interaction for th jm ne lines studied during both 2006 and 2007 seasons (7 lines out of a total 33 lines) is presented in Addendum 8 and in Figure 4.9.4.

Figure 4.9.4. Biplot of the starch yield G×E interaction analysis, combined 2006-2007 seasons



The top-3 high starch yielding genotypes estimated from the combined 2006–2007 seasons AMMI2 model were YC, CA and D1 (Addendum 8; Table 4.9.7).

Table 4.9.7. The best genotypes of the 2006-2007 seasons for starch yield suggested by the AMMI2 model with high yield potential and adaptation
(values fitted for G×E interaction, kg.ha^{-1})

Location	Swartland					Overberg				
Score	VR	PI	KL	LA	ME	RO	TY	NA	RI	AL
2006 season										
1st	**CD** 2242			**CD** 2064	**YC** 1622	**CA** 3378	**Y2** 2387	**CA** 3166		
2nd	**YC** 1964			**CA** 2021	**CA** 1574	**YC** 3367	**YC** 2365	**Y2** 3113		
2007 season										
1st		**CA** 3092	**CD** 3052	**CA** 3625	**Y2** 2227	**YC** 3898	**Y2** 3293	**Y2** 3756	**CE** 3160	**YC** 3521
2nd		**YC** 3064	**YC** 2799	**Y2** 3475	**CC** 2186	**CA** 3889	**D1** 3229	**CC** 3406	**YC** 3122	**CC** 3467

For the Swartland region high starch yielding lines YC, CA and CD could be recommended, and for the Overberg lines Y2, YC and CA. However, taking into account small number of lines assessed during both seasons the recommendation of these lines cannot be viewed as final. Other lines which showed high performance during one of the seasons and were not assessed in another season trials are strongly recommended to be considered for re-evaluation.

4.10. ETHANOL OUTPUT, ETHANOL YIELD, AAQ AND TEST WEIGHT ANALYSES, THE 2007 SEASON

Ethanol output (L.tonne^{-1}) with (+E) and without (-E) addition of technical saccharifying enzymes, autoamylolytical quotient (AAQ, %) and ethanol yield (L.ha^{-1}) data are presented in Tables 4.10.1 – 4.10.5. Results for AAQ were in disagreement with results of other research studies found in literature – much higher AAQ values were expected (THOMAS *et al.*, 1991; SENN *et al.*, 1993; SENN & PIEPER, 2001). The disagreement can be explained by some differences in fermentation protocols used to assess the material under investigation, particularly elimination of a high-temperature jet-cooking step in our study, which is not required by modern technical enzymes *viz*. STARGEN™ 002 used. Another reason could be a lower pH during gelatinisation and fermentation stages in our study (pH = 3.6 against recommended 5.0–5.8; see THOMAS *et al.*, 1991; SENN *et al.*, 1993). Such low pH level is not optimal for endogenous enzymes; hence their activity could be hindered. Yet another reasons (however less probable) might be that endogenous enzymes of the lines under investigation were indeed lacking much of expected substantial natural activity (or level of enzyme inhibitors was high), thus leading to low AAQ levels.

4.10.1. Ethanol output analysis

The ethanol output ANOVA has shown that the difference between replicates of the fermentations was greater than the difference between genotypes (Table 4.10.1.1). Only some trial locations were statistically distinguishable from each other by the level of ethanol output (figures are not shown). Ranking of the results has shown that line D3 and location Langgewens were characterised by the highest mean ethanol output, and the line EB and locations Mariendahl, Napier and Riversdale were characterised by lowest ethanol output (Addendum 9; Table 4.10.1.2).

Table 4.10.1.1. ANOVA of ethanol output results of the 2007 season

	SS	DF	MS	F-ratio	p-value
Summary					
Model	8.88E+05	27	3.29E+04	23.673	0.000
Error	7.11E+05	512	1.39E+03		
Adjusted Total	1.60E+06	539	2.97E+03		
Variable:					
Entry	2.75E+04	19	1.45E+03	1.04	0.412
Location	8.60E+05	8	1.08E+05	77.426	0.000

Table 4.10.1.2. ANOVA and ranking indices of the 2007 season ethanol output across locations

(A) ANOVA

Source	DF	SS	MS	F-ratio	P-value
Entry	19	6741.45	354.81	0.97594	1
Location	8	211319.10	26414.89	72.65638	0.00001
Interaction (Error)	152	55260.98	363.56		

Other statistic parameters:

Locations	9	**LSD(5%)**	18.156
Mean	471.627	**Pairwise SE**	8.988
Std Error (SE)	19.067	**CV%**	4.04
Std Deviation (SD)	6.279	**Mean/LSD**	25.976
Vp	39.426	**H^2=Vg/Vp**	-0.025
Vg	-0.97	**G/GGE% (SS)**	10.87
MSe	363.559	**G/GGE% (VC)**	-0.27

Table 4.10.1.2. (continued)
(B) Ranking indices*

Entry	Mean EO, L.tonne^{-1}	RV, %	HARV, %	SI, %	HASI, %	Mean/ LSD5%	Mean/ LSD1%
D3	483.03	102	100	100	100	26.60	20.14
H1	478.67	101	100	99	100	26.36	19.96
G2	477.31	101	100	99	100	26.29	19.90
EA	476.76	101	100	99	100	26.26	19.88
D2	475.89	101	100	99	100	26.21	19.84
D1	474.84	101	100	98	100	26.15	19.80
G1	474.79	101	100	98	100	26.15	19.80
CC	474.23	101	100	98	100	26.12	19.77
H4	474.18	101	100	98	100	26.12	19.77
H2	473.30	100	100	98	100	26.07	19.74
CE	473.22	100	100	98	100	26.06	19.73
H6	472.05	100	100	98	100	26.00	19.68
CA	471.78	100	100	98	100	25.98	19.67
H3	468.66	99	100	97	100	25.81	19.54
D4	467.65	99	100	97	100	25.76	19.50
YC	466.50	99	100	97	100	25.69	19.45
Y2	466.39	99	100	97	100	25.69	19.45
H5	464.77	99	100	96	100	25.60	19.38
CD	462.60	98	100	96	100	25.48	19.29
EB	455.93	97	100	94	100	25.11	19.01

* See remarks for Table 4.9.5.

As can be seen from the Table 4.10.1.2-B, mean ethanol output of the top-17 lines was ranging between 466–483L.tonne^{-1}, with the 95%CI ethanol output range being 466–477L.tonne^{-1} (Table 4.9.1). These results are relatively high compared to such reported for triticale and wheat in the literature, e.g. 370–460L.tonne^{-1} (DM) of triticale grain with addition of technical enzymes (KUCEROVA, 2006), average of 436L.tonne^{-1} (DM) of triticale grain without addition of industrial enzymes (DAVIS-KNIGHT & WEIGHTMAN, 2008), up to 465L.tonne^{-1} (DM) of wheat grain (ROSENBERGER, 2005), and 410–480L.tonne^{-1} of wheat grain (SMITH *et al*., 2006). It could be concluded that selection for higher ethanol output would be difficult because of the low variation of the trait among the investigated triticale lines. More lines are needed to be evaluated for the trait in subsequent seasons.

Results of the AMMI2 model analysis (Addendum 9 and Figure 4.10.1.1) revealed that lines G2, D1, CD, D3, H4 and H6 possess high levels of the G×E

interactions for ethanol output. The best lines for ethanol output for each location are presented in Table 4.10.1.3 and Figure 4.10.1.2, namely G2, D3 and H2 for the Swartland region and D3, G2 and D1 for the Overberg.

Figure 4.10.1.1. Two-way interaction biplot of the AMMI2 model for the 2007 season ethanol output

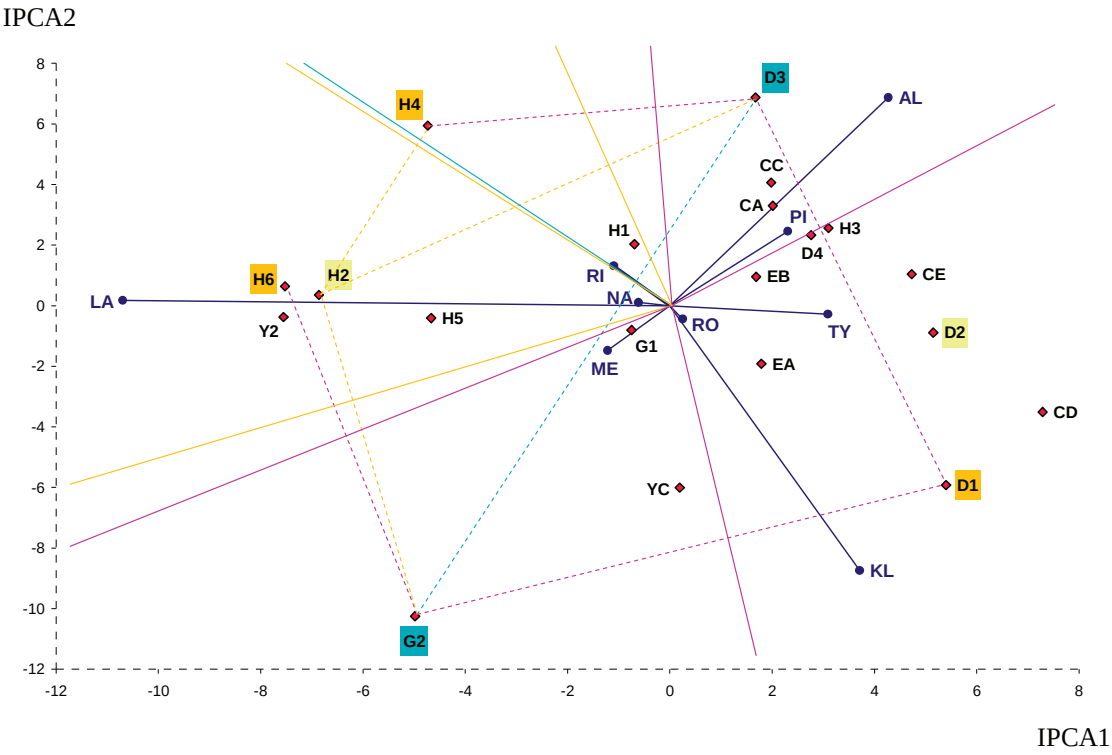
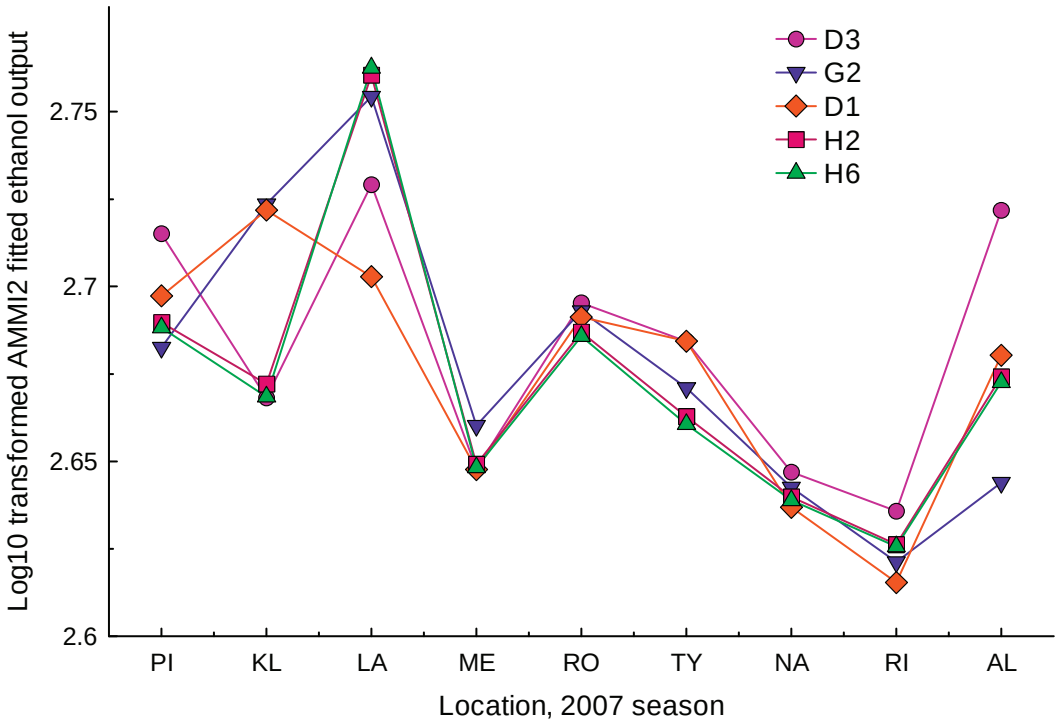


Table 4.10.1.3. The best genotypes across the 2007 season locations for ethanol output with high stability of the output potential

(L.tonne^{-1} fitted for the G×E interaction)

Location	Swartland				Overberg				
Best genotype	**PI**	**KL**	**LA**	**ME**	**RO**	**TY**	**NA**	**RI**	**AL**
1st	**D3** 518.9	**G2** 529.2	**H6** 578.8	**G2** 457.3	**D3** 495.8	**D1** 483.5	**D3** 443.6	**D3** 432.3	**D3** 527.0
2nd	**D2** 505.7	**D1** 527.1	**H2** 576.1	**H2** 445.9	**G2** 493.2	**D3** 483.4	**G2** 439.1	**H4** 426.6	-
3rd	-	-	**G2** 568.0	**H6** 444.9	**D1** 491.2	**D2** 483.4	**H2** 436.4	-	-

Figure 4.10.1.2. Comparison of the best five triticale lines suggested by AMMI2 model for ethanol output across locations of the 2007 season



Calculations of theoretical ethanol output values (based on total starch content) and their comparison to actual ethanol output, termed here as relative ethanol output (REO, %) are given in Table 4.10.1.4. It can be seen that in some locations majority of lines gave higher than theoretical ethanol output, while in other locations their ethanol output was lower than expected from theoretical values. Such results in locations with higher than theoretical ethanol output can be explained by a reason that some NSP (see section 2.3 “Non-starch polysaccharides”), as well as portions of cellulose and hemicellulose were successfully digested into fermentable sugars by technical enzymes (OPTIMASH™ VR and STARGEN™ 002). Consequently, extra portion of these fermentable sugars could be converted into ethanol by yeast during SSF while being not accounted for by the total starch assay.

Table 4.10.1.4. Relative ethanol output (REO), % of theoretical values calculated from starch content

Entry	Line code name	Location									Mean	St. Dev.
		PI	LA	KL	ME	RO	TY	NA	RI	AL		
1	**CA**	99.75	105.31	110.00	91.71	109.04	109.56	94.24	95.69	112.98	103.14	7.46
2	**CC**	110.10	99.24	112.64	95.57	108.17	112.81	93.48	97.27	115.75	105.00	8.07
3	**CE**	107.23	108.46	110.11	95.16	103.92	102.96	91.88	94.91	120.09	103.86	8.40
4	**CD**	105.42	111.98	105.69	95.04	89.73	107.80	94.03	94.91	106.99	101.29	7.39
5	**D1**	101.57	116.14	108.94	96.27	107.19	106.36	94.06	95.34	107.45	103.70	6.98
6	**D2**	109.15	106.68	109.86	92.52	102.37	111.85	92.61	93.05	109.96	103.12	7.76
7	**D3**	109.59	102.25	115.46	95.62	102.26	104.58	90.04	95.12	120.62	103.95	9.36
8	**D4**	108.71	100.92	108.48	92.95	103.51	98.42	89.74	96.37	112.14	101.25	7.19
9	**YC**	102.30	113.15	114.95	97.60	105.39	110.67	89.30	95.73	104.00	103.68	8.01
10	**Y2**	104.18	101.72	118.76	95.75	102.28	105.19	94.71	94.34	102.53	102.16	7.05
11	**EA**	107.11	108.44	110.48	92.66	102.31	102.55	97.49	97.56	114.10	103.63	6.58
12	**EB**	101.10	100.31	107.87	96.64	98.81	106.74	93.25	92.85	113.89	101.27	6.64
13	**G1**	96.28	105.54	113.70	91.99	106.24	98.81	91.63	92.89	111.35	100.94	8.02
14	**G2**	100.77	115.81	119.59	98.96	106.33	102.06	92.81	92.05	96.59	102.78	9.05
15	**H1**	107.65	104.07	115.72	91.05	104.97	109.68	96.98	97.46	109.48	104.12	7.25
16	**H2**	102.33	101.39	121.50	94.95	102.24	105.53	92.85	95.83	104.18	102.31	7.95
17	**H3**	102.69	97.83	102.42	94.36	102.53	105.40	96.09	92.79	103.57	99.74	4.28
18	**H4**	111.35	97.28	122.70	93.87	100.95	102.68	91.01	93.82	113.10	102.97	10.04
19	**H5**	98.68	100.55	116.42	97.20	101.42	104.22	94.73	97.50	106.56	101.92	6.18
20	**H6**	102.41	99.45	120.01	89.98	103.19	99.60	91.94	92.36	101.51	100.05	8.45
Mean (site index)		104.42	104.83	113.27	94.49	103.14	105.37	93.14	94.89	109.34		
Standard Deviation		4.15	5.69	5.40	2.30	3.97	4.06	2.20	1.77	6.09		

On the other hand, lower than theoretical ethanol output in some locations is probably due to an environmental factor that affected starch and NSP structure or their composition in kernels, which could made it more difficult for enzymes to digest the substrate (TESTER & SOMMERVILLE, 2003; JOBLING, 2004; BRENNAN & CLEARY, 2005). It is known that fractional composition and technological properties of majority of starches and other polysaccharides significantly depend on soil-climatic growth conditions (MYLLÄRINEN *et al.*, 1998; DEBON *et al.*, 1998; TESTER & KARKALAS, 2001). This in turn could hinder availability of free sugars to yeast, thus limiting ethanol output from carbohydrates. Measurement of residue viscosities could be used in future research for an indirect estimation of grain quality for ethanol production, because high-ethanol yielding cultivars tend to give low residue viscosities, which can be explained by a negative correlation between NSP and starch content of whole grain (SMITH *et al.*, 2006).

Besides the total starch content *per se*, it seems possible that a change in the ratio of starch components (i.e. amylose and amylopectin) and granule morphology and size, its extent of crystallinity or damage could also altered kernel processing characteristics thus influenced ethanol output from grain (THEMEIER *et al.*, 2005; RUDI *et al.*, 2006; SAJILATA, SINGHAL & KULKARNI, 2006). Large A-type granules were shown to be easily hydrolysed by amylases because they have loosely packed internal structures, in contrast to smaller B-type granules (JANE *et al.*, 2003). However, it was found that total starch content (sum of A and B type granules) is more important for ethanol yield than relative amounts of starch granules of different type (BROSNAN *et al.*, 1999). Difference in hardness of grain harvested from different locations could also affect ethanol output because of its influence on particle size, which results from different milling properties of harder and softer grain (see

discussion of ethanol output and ethanol yield in section 4.9 “Combined analysis of the 2007 season data” for more information). The difference between fine and coarser ground meal may be as much as 5–10% of ethanol output (KELSALL & LYONS, 2003). Other research studies have found that ethanol exploitation increased when grain was harvested few weeks later than at the stage of full ripeness (AUFHAMMER, 1998), which could activate endogenous enzymes. The abovementioned reasons could also have an effect in our research.

The bioavailability of starch may differ among cereal cultivars and may affect the conversion rate and final yield of ethanol (MOORTHY, 2002; ROSENBERGER, 2005). From the further analysis of the REO data presented in the Addendum 10 and summarised in Figure 4.10.1.3 it is evident that some triticale lines had higher G×E interaction than others for the trait. Genotypes CC, D3, CE, D1, H4, G2 and H2 were best performers for the REO. Comparison of the REO with actual ethanol output (Addendum 9 and Figure 4.10.1.1) and total starch content (Addendum 5) leads to conclusion that performance of analysed lines in given locations in terms of ethanol output can only be partially explained by their total starch content (correlation of ethanol output with total starch content was $R^2 = 0.359$, $P < 0.001$; Table 4.9.3). Another influential variable was the bioavailability (i.e. easiness of accessibility and digestibility) of starch and other polysaccharides to enzymes and yeast, which is described by the REO (correlation of actual ethanol output with REO was $R^2 = 0.898$, $P < 0.001$; see Table 4.10.1.5).

Considering the above, ethanol output is shown to be more dependent on starch and other polysaccharides accessibility to enzymatic digestion than on the total starch content as such. Thus, the total starch content alone taken as a possible indirect predictor of the ethanol output without consideration of the actual ethanol output can

lead to incorrect conclusions, considering their low correlation (R^2 = 0.359, P <0.001; Table 4.9.3). Because the small-scale fermentation is relatively easy and inexpensive to perform compared to determination of total starch or protein content, as well as other grain parameters linked to ethanol output, the question "what causes one cultivar to give higher ethanol outputs than the other?" seems to be of a lesser importance from practical point of view. The more important question in this stage of the research might be "would the results for ethanol output obtained in small-scale fermentation process correspond with the results of a large-scale industrial process?" Therefore, realisation of larger-scale fermentation alongside with small-scale fermentation and comparison of their results would be necessary in the future research.

Figure 4.10.1.3. Two-way interaction biplot of the AMMI2 model for the 2007 season for relative ethanol output (REO)

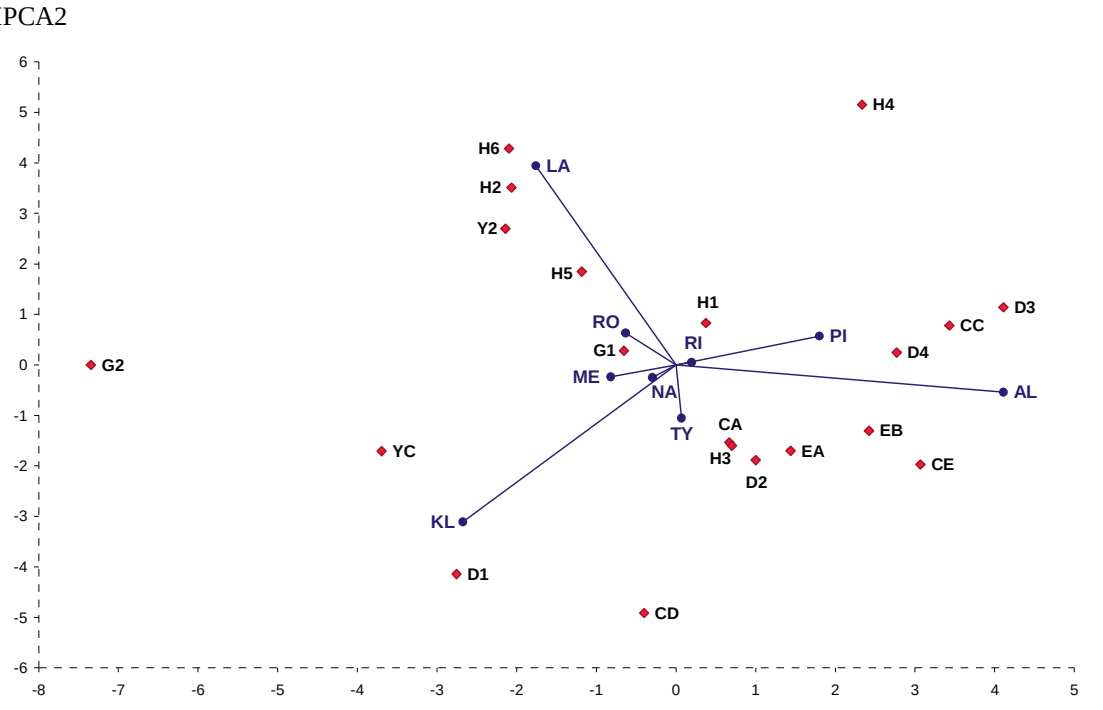


Table 4.10.1.5. Spearman' rank correlation (SRC) of relative ethanol output (REO) with other traits†

REO, %	Days to heading	Height, cm	Test weight, kg.HL^{-1}	PSI, %	Protein content, %	Moisture content, %
SRC	–0.166	–0.369	–0.046	–2.61E-04	–0.017	0.547
t-test value	–2.225	–4.649	–0.618	–0.003	–0.221	7.318
Significance level	*	***	N.S.	N.S.	N.S.	***
Probability > t	0.026	3.34E-06	0.537	0.997	0.825	2.52E-13

REO, %	Starch content, %	Grain yield as-is, kg.ha^{-1}	Starch yield, kg.ha^{-1}	Ethanol output, L.tonne^{-1}	Ethanol yield, L.ha^{-1}
SRC	–0.066	0.231	0.217	0.898	0.571
t-test value	–0.881	3.084	2.902	12.012	7.639
Significance level	N.S.	**	**	***	***
Probability > t	0.378	0.002	0.004	3.09E-33	2.18E-14

† Significance levels as described in Table 4.9.3.

4.10.2. Ethanol yield analysis

Results of the ethanol yield ANOVA and ranking indices are given in Table 4.10.2.1, and the AMMI2 model of the cross-site analysis of the trait is presented in the Addendum 11 and Figure 4.10.2.1. The 95%CI of the ethanol yield for all lines ranged between 2446–2625L.ha^{-1} (Table 4.9.1), and mean ethanol yield of the top-15 lines was in the range of 2510–2787L.ha^{-1}; broad-sense heritability $H^2 = 0.824$. Ethanol yield can be described as having wider variability and higher heritability compared to ethanol output thus selection for this trait might be more successful.

Table 4.10.2.1. The ANOVA and ranking indices of the 2007 season ethanol yield across locations

(A) ANOVA

Source	DF	SS	MS	F-ratio	P-value
Entry	19	9470740	498460	5.6814	0.00001
Location	8	43350082	5418760	61.7621	0.00001
Interaction (Error)	152	13335873	87736		

Other statistic parameters:

Locations	9	**LSD(5%)**	282.05
Mean	2535.714	**Pairwise SE**	139.631
Std Error (SE)	296.203	**CV%**	11.68
Std Deviation (SD)	235.338	**Mean/LSD**	8.99
Vp	55383.97	**H^2=Vg/Vp**	0.824
Vg	45635.53	**G/GGE% (SS)**	41.53
MSe	87736.01	**G/GGE% (VC)**	34.22

Table 4.10.2.1. (continued)
(B) Ranking indices*

Entry	Mean ethanol yield, L.ha^{-1}	RV, %	HARV, %	SI, %	HASI, %	Mean/ LSD5%	Mean/ LSD1%
G2	2786.53	110	108	100	100	9.88	7.48
D2	2735.61	108	107	98	98	9.70	7.34
D1	2702.67	107	106	97	98	9.58	7.25
G1	2690.66	106	105	97	98	9.54	7.22
D4	2677.74	106	105	96	97	9.49	7.19
H1	2669.23	105	104	96	97	9.46	7.16
D3	2655.11	105	104	95	96	9.41	7.13
H3	2637.35	104	103	95	96	9.35	7.08
EA	2632.38	104	103	94	95	9.33	7.07
YC	2620.97	103	102	94	95	9.29	7.04
H2	2603.68	103	102	93	94	9.23	6.99
H6	2590.31	102	102	93	94	9.18	6.95
CC	2587.07	102	102	93	94	9.17	6.94
Y2	2544.69	100	100	91	93	9.02	6.83
CA	2510.16	99	99	90	92	8.90	6.74
H4	2472.41	98	98	89	91	8.77	6.64
CE	2456.80	97	98	88	90	8.71	6.59
EB	2186.47	86	88	78	82	7.75	5.87
H5	2137.47	84	87	77	81	7.58	5.74
CD	1816.97	72	77	65	71	6.44	4.88

* See remarks for Table 4.9.5.

Bio-ethanol yield per hectare is demonstrated to be largely a function of grain yield, thus it depends on agronomic intensity level that is mainly determined by a level of nitrogen supply (TAYLOR & ROSCROW, 1990; SMITH *et al.*, 2006). Variation in ethanol yield between sites and years is commonly larger than between cultivars (SMITH *et al.*, 2006). Because different genetic mechanisms influence ethanol yield, it could be possible to develop improved breeding lines by combining desired ethanol yield traits from complementary parents (SWANSTON *et al.*, 2007). The best ethanol yielding lines for each location were selected by analysis of the Addendum 11 and Figure 4.10.2.1 and are presented in Table 4.10.2.2. The most unstable genotype across locations with the highest G×E interaction was CD, and on the contrary lines D2 and D1 were most stable in terms of ethanol yield across locations. For the Swartland region the best genotypes were D1, H1 and D2, and for the Overberg

genotypes H1 and G2. Graphical comparison of these four best genotypes is presented in Figure 4.10.2.2.

Figure 4.10.2.1. Two-way interaction biplot of the AMMI2 model for the 2007 season ethanol yield

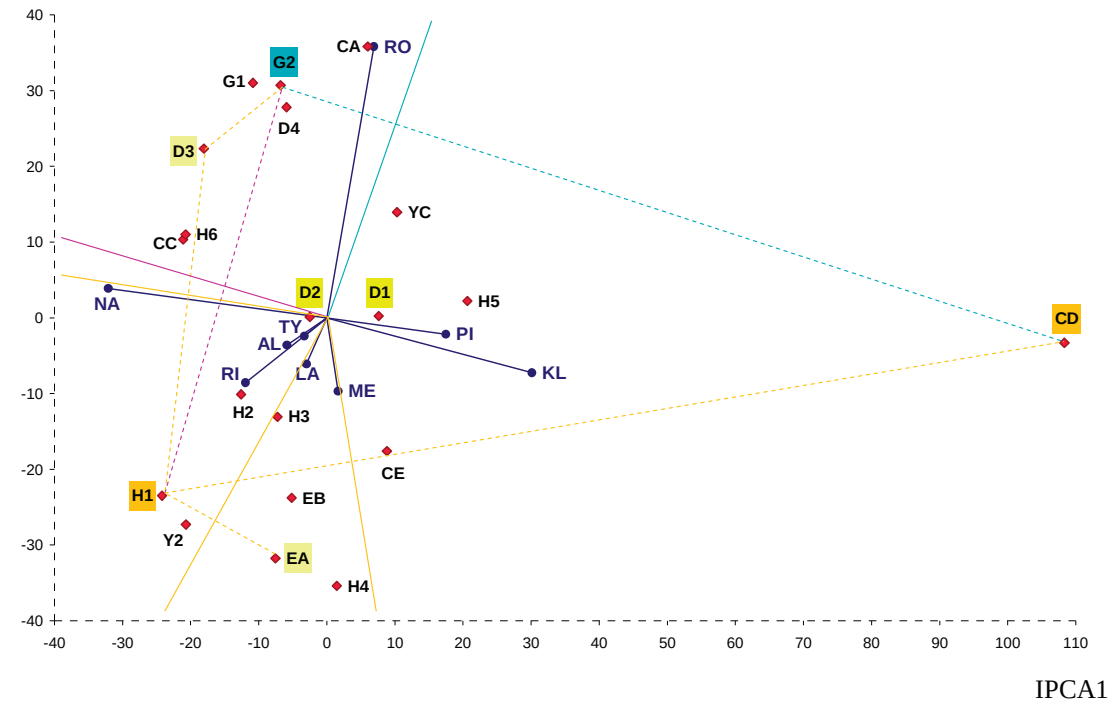
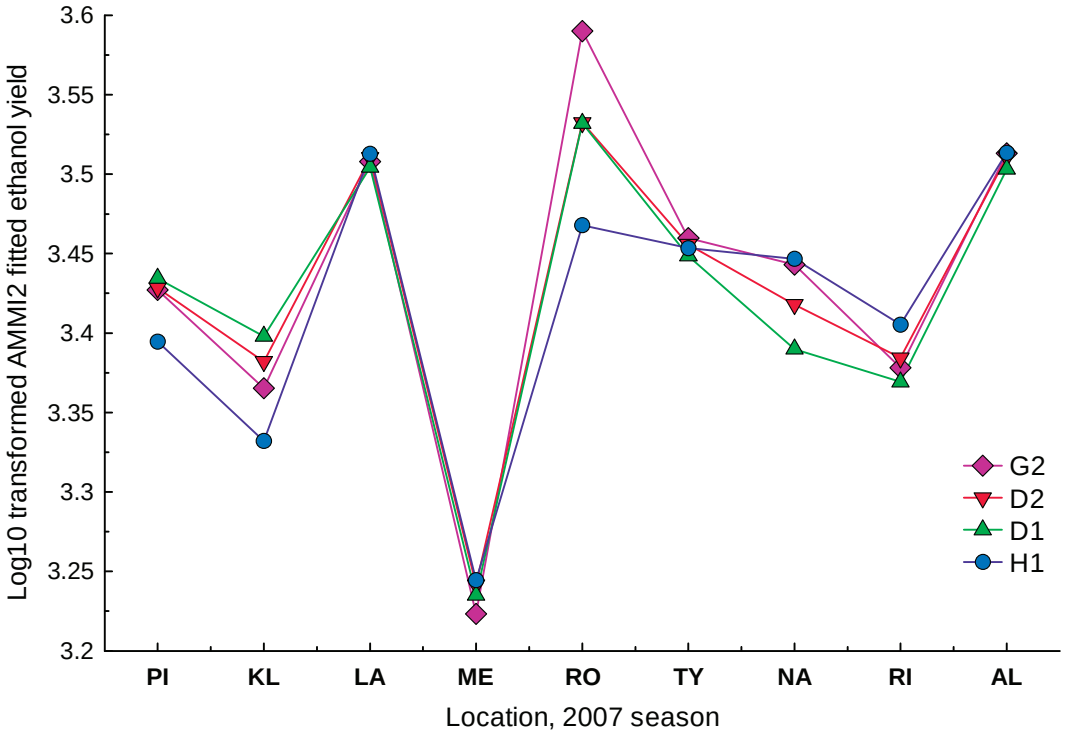


Table 4.10.2.2. The best genotypes across the 2007 season locations for ethanol yield with high stability of yield potential

(kg.ha^{-1} fitted for G×E interaction)

Location	Swartland				Overberg				
Score	**PI**	**KL**	**LA**	**ME**	**RO**	**TY**	**NA**	**RI**	**AL**
1st	**D1** 2719	**CD** 2855	**H1** 3257	**EA** 1763	**G2** 3890	**G2** 2883	**H1** 2797	**H1** 2543	**H1** 3262
2nd	**D2** 2681	**D1** 2500	**D2** 3239	**H1** 1756	**D3** 3605	**D2** 2856	**D3** 2775	**EA** 2454	**G2** 3259
3rd	**G2** 2674	**D2** 2410	**EA** 3221	**D2** 1744	**D2** 3407	**H1** 2841	**G2** 2774	**D2** 2423	**D2** 3242

Figure 4.10.2.2. Comparison of the best four triticale lines for ethanol yield across locations for the 2007 season, suggested by the AMMI2 model



Considering Figure 4.9.3 and Table 4.10.2.2, breeding combinations D1 / H1, CD / D1, CD / D2 and CD / H1 for the Swartland region, and H1 / G2 for the Overberg region, as well as D2 / H1 and D2 / G2 for both sub-environments could be recommended for the achievement of an increased ethanol yield.

4.10.3. Test weight analysis

Test weight is an important trait which influences grain yield, thus was also expected to positively influence ethanol yield (ANONYMOUS, 2003; JACOBI & HARTMANN, 2005). However, positive correlation between these traits was weak ($R^2 = 0.279$, $P < 0.001$ for test weight and grain yield correlation, and $R^2 = 0.238$, $P < 0.01$ for test weight and ethanol yield correlation; see Table 4.9.3). The ANOVA for the genotypes across locations and their ranking is summarised in Table 4.10.3.1. Results of the G×E interaction AMMI2 analysis of test weight are presented in

Addendum 12. The best lines in terms of test weight were EB and Y2. However, they were the ones on the lower end for ethanol yield (Table 4.10.2.1), as well as for ethanol output (Table 4.10.1.2). The trait did not show any significant correlation with the REO (Table 4.10.1.5). Considering the abovementioned results, test weight cannot be used as a reliable predictor of genotypes performance for ethanol output and ethanol yield.

Table 4.10.3.1. ANOVA and ranking indices of the 2007 season test weight across locations

(A) ANOVA

Source	DF	SS	MS	F	P
Entry	19	258.0799	13.58315	7.756884	0.00001
Location	8	317.0255	39.62818	22.63033	0.00001
Interaction (Error)	152	266.1686	1.75111		

Other statistic parameters:

Locations	9	**LSD(5%)**	1.26
Mean	73.461	**Pairwise SE**	0.624
Std Error (SE)	1.323	**CV%**	1.8
Std Deviation (SD)	1.228	**Mean/LSD**	58.3
Vp	1.508	**H^2=Vg/Vp**	0.871
Vg	1.313	**G/GGE% (SS)**	49.23
MSe	1.751	**G/GGE% (VC)**	42.85

(B) Ranking indices (see remarks for Table 4.9.5)

Entry	Mean test weight, kg.HL^{-1}	RV, %	HARV, %	SI, %	HASI, %	Mean/ LSD5%	Mean/ LSD1%
EB	75.85	103	103	100	100	60.20	45.57
Y2	75.70	103	103	100	100	60.08	45.48
EA	74.67	102	102	98	98	59.26	44.86
H4	74.59	102	102	98	98	59.20	44.82
G1	74.44	101	101	98	98	59.08	44.73
H3	74.22	101	101	98	98	58.90	44.59
G2	73.93	101	101	97	97	58.67	44.42
H6	73.85	101	101	97	97	58.61	44.37
H5	73.48	100	100	97	97	58.32	44.15
H1	73.41	100	100	97	97	58.26	44.10
YC	73.33	100	100	97	97	58.20	44.06
D3	73.26	100	100	97	97	58.14	44.01
CA	73.11	100	100	96	97	58.02	43.93
H2	72.89	99	99	96	97	57.85	43.79
D2	72.52	99	99	96	97	57.55	43.57
D1	72.44	99	99	96	97	57.49	43.53
CD	72.41	99	99	95	96	57.46	43.50
CE	72.15	98	98	95	96	57.26	43.35
D4	71.78	98	98	95	96	56.96	43.12
CC	71.19	97	97	94	95	56.49	42.77

4.11. RESULTS OF THE NIRS PREDICTION MODELS DEVELOPMENT

Near infra-red reflectance spectra data of triticale samples from the 2006 and 2007 seasons are presented in Tables A4.11.1 – A4.11.4 (Addendum 13); corresponding data for moisture (%) and starch content (% 'as-is' and on DWB) are also included. Chemometric analysis of NIRS data with moisture and total starch content data was performed and prediction models for these traits were developed for the 2006 and 2007 season's datasets.

4.11.1. Calibrations results for the 2006 season

Results of the study for the 2006 season were partially reported in a short communication (Y. Tsupko, W.C. Botes, M. Manley. Development of near infra-red reflectance spectroscopy calibration models for starch and moisture content in ×*Triticosecale* Wittmack. In press). Complete results for the 2006 season are presented in Table 4.11.1.1 for whole kernels and in Table 4.11.1.2 for milled triticale grain samples.

In the 2006 season, 200 lines from elite and senior trials were assessed for total starch and moisture content. The traits ranged between 49.46–61.92% (mean = 55.48%, SD = 2.096) for the total starch ('as-is') and 9.51–11.81% (mean = 10.77%, SD = 0.453) for moisture content. The PLS1 models based on whole grain spectra did not show promising outcomes (Table 4.11.1.1). For moisture content the RPD ranged between 1.107–1.253 units for the complete dataset with the best squared coefficient of determination $R^2 = 0.364$ achieved with the MSC + 2nd derivative data pre-treatment. When the dataset was cleaned from outliers, the best RPD was 1.320 units with the best pre-treatment being the MSC with achieved $R^2 = 0.441$ and SEP = 0.333%. For starch content ('as-is') the best RPD's were 1.091

(complete dataset) and 1.095 (without outliers), with $R^2 = 0.2$. Results for starch content (DWB) were slightly better with the RPD for complete dataset ranging between 1.014–1.118, the best being $R^2 = 0.214$, and developed from dataset cleaned from outliers the best pre-treatment was MSC with the RPD = 1.145 and $R^2 = 0.25$.

Table 4.11.1.1. Cross-validation prediction results of the PLS1 calibration models for starch and moisture content in triticale whole kernels, the 2006 season

Statistics	Spectral data pre-treatment					
	Raw spectra	MSC	MSC + 1st Der.	MSC + 2nd Der.	1st Der.	2nd Der.
Moisture content, complete dataset						
N	200	200	200	200	200	200
Mean, %	10.770	10.770	10.770	10.770	10.770	10.770
St.Dev.	0.453	0.453	0.453	0.453	0.453	0.453
PLS factors	7	8	3	2	3	2
Slope	0.316218	0.361152	0.277861	0.386254	0.361539	0.399230
R^2 validation	0.241594	0.271879	0.206933	0.364099	0.294320	0.345082
RMSEP, %	0.399218	0.393033	0.408299	0.361528	0.383638	0.368533
SEP, %	0.400213	0.394015	0.409309	0.361405	0.384429	0.368750
Bias	–0.002200	–0.001932	–0.003482	–0.027236	–0.011448	–0.022791
RPD	1.132	1.150	1.107	1.253	1.178	1.228
Moisture content, without outliers						
N	198	197	198	198	198	199
Mean, %	10.774	10.768	10.771	10.780	10.771	10.776
St.Dev.	0.453	0.439	0.440	0.443	0.440	0.445
PLS factors	13	10	3	2	3	2
Slope	0.495207	0.523302	0.324515	0.368393	0.396413	0.417212
R^2 validation	0.360532	0.440771	0.256987	0.376640	0.338253	0.366169
RMSEP, %	0.377052	0.332355	0.382923	0.348889	0.359638	0.355745
SEP, %	0.376567	0.332675	0.383671	0.349550	0.360528	0.356190
Bias	–0.032891	0.018682	–0.013057	–0.012460	–0.004005	–0.017904
RPD	1.203	1.320	1.147	1.267	1.220	1.249

(continued)

Table 4.11.1.1. (continued)

Statistics	Spectral data pre-treatment					
	Raw spectra	MSC	MSC + 1st Der.	MSC + 2nd Der.	1st Der.	2nd Der.
Starch content as-is, complete dataset						
N	200	200	200	200	200	200
Mean, %	55.481	55.481	55.481	55.481	55.481	55.481
St.Dev.	2.096	2.096	2.096	2.096	2.096	2.096
PLS factors	9	7	4	2	5	3
Slope	0.299905	0.247609	0.263664	0.168534	0.275801	0.179644
R^2 validation	0.204037	0.165874	0.181694	0.100519	0.156871	0.109378
RMSEP, %	1.919840	1.955038	1.933755	2.032859	2.022742	2.022499
SEP, %	1.922040	1.959828	1.938285	2.037951	2.024748	2.027426
Bias	0.100098	–0.021255	0.035239	0.006260	0.111270	0.024439
RPD	1.091	1.069	1.081	1.028	1.035	1.034
Starch content as-is, without outliers						
N	197	198	199	199	198	199
Mean, %	55.569	55.421	55.449	55.458	55.431	55.458
St.Dev.	1.987	2.018	2.050	2.075	2.043	2.075
PLS factors	9	7	4	2	5	3
Slope	0.294016	0.249151	0.264089	0.190213	0.296481	0.198524
R^2 validation	0.204680	0.177196	0.188908	0.121485	0.172999	0.133347
RMSEP, %	1.812383	1.858085	1.875918	1.982688	1.950914	1.962148
SEP, %	1.815359	1.862752	1.880411	1.987663	1.954815	1.967096
Bias	0.077011	–0.012532	–0.029820	0.010121	0.063763	0.001831
RPD	1.095	1.083	1.090	1.044	1.045	1.055
Starch content DWB, complete dataset						
N	200	200	200	200	200	200
Mean, %	62.181	62.181	62.181	62.181	62.181	62.181
St.Dev.	2.389	2.389	2.389	2.389	2.389	2.389
PLS factors	7	6	4	2	2	2
Slope	0.272059	0.268652	0.236243	0.193645	0.155473	0.182024
R^2 validation	0.204970	0.214337	0.133419	0.123781	0.085298	0.122544
RMSEP, %	2.155236	2.131148	2.318209	2.281150	2.351068	2.269719
SEP, %	2.159803	2.136465	2.323868	2.286500	2.356331	2.274826
Bias	–0.060148	–0.011491	0.027092	0.041269	–0.054649	0.051627
RPD	1.106	1.118	1.028	1.045	1.014	1.050
Starch content DWB, without outliers						
N	198	198	198	199	198	199
Mean, %	62.231	62.123	62.124	62.157	62.124	62.157
St.Dev.	2.347	2.329	2.332	2.371	2.332	2.371
PLS factors	8	7	5	1	4	2
Slope	0.299015	0.307273	0.330693	0.169427	0.248077	0.177975
R^2 validation	0.229706	0.250226	0.240417	0.120455	0.167583	0.128482
RMSEP, %	2.083488	2.028974	2.072518	2.243365	2.171375	2.234231
SEP, %	2.087815	2.033772	2.077596	2.248933	2.176814	2.237814
Bias	–0.062972	–0.037383	–0.026980	–0.019954	–0.016770	–0.095605
RPD	1.124	1.145	1.122	1.054	1.071	1.060

Calibrations development done for milled grain samples resulted in generally better outcomes, especially for moisture content, however all calibrations being still

weak in relative prediction power if had to be used for practical applications (Table 4.11.1.2). The best calibration for moisture content after the MSC + 1st derivative data pre-treatment had the RPD = 2.333 with R^2 = 0.816, SEP = 0.191% and 3 PLS factors. Calibration with such parameters is suitable for a rough screening (ranking) of samples (WILLIAMS, 2001).

Calibrations for starch content ('as-is') developed from a complete dataset produced best results with the MSC + 1st derivative data pre-treatment (RPD = 1.304, R^2 = 0.43) and for a dataset without outliers the 1st derivative alone data pre-treatment gave the RPD = 1.346, R^2 = 0.46 with SEP = 1.507% and 4 PLS factors. Calibrations for starch content on DWB resulted in comparable results (see Table 4.11.1.2).

Results of the 2006 season calibrations have shown that the MSC, MSC + 1st derivative and 1st derivative alone were the most fruitful spectral data pre-treatments both for whole kernels and for milled grain samples. Datasets cleaned from explicit outliers always resulted in more robust calibrations with better predictive power. However, general quality of the calibrations (even the best ones) was low (WILLIAMS, 2001), especially for starch content. It can be explained by a poor initial data quality for moisture content as well as for starch content, relatively small number of reference samples (*ca*. five to ten fold smaller compared to reported by other researches; see sections 2.7.4 "Instrumental grain composition measurements – near infra-red reflectance/ transmittance (NIR/NIT) spectroscopy" and 2.7.4.1 "Near infra-red spectroscopy application examples – starch and other constituents determination") and also by a general difficulty of calibrations development for starch content. The difficulty rises from the complexity and non-uniformity of starchy compounds, with the best calibrations described in scientific literature characterised by the RPD in the range of 2–3 units being common for starch measurements (PAULSEN & SINGH, 2004).

Table 4.11.1.2. Cross-validation prediction results of the PLS1 calibration models with different data pre-treatments for starch and moisture contents in triticale milled grain, the 2006 season

Statistics	Spectral data pre-treatment					
	Raw spectra	MSC	MSC + 1st Der.	MSC + 2nd Der.	1st Der.	2nd Der.
Moisture content, complete dataset						
N	200	200	200	200	200	200
Mean, %	10.770	10.770	10.770	10.770	10.770	10.770
St.Dev.	0.453	0.453	0.453	0.453	0.453	0.453
PLS factors	5	4	2	3	3	3
Slope	0.768314	0.757615	0.732543	0.643199	0.740691	0.624599
R^2 validation	0.754740	0.753540	0.739939	0.701239	0.746842	0.693436
RMSEP, %	0.223759	0.224204	0.230333	0.248843	0.227287	0.253001
SEP, %	0.224321	0.224767	0.230908	0.249440	0.227813	0.253447
Bias	–0.000315	0.000187	–0.001194	0.003678	–0.004455	0.009767
RPD	2.019	2.015	1.962	1.816	1.988	1.787
Moisture content, without outliers						
N	197	197	198	198	197	198
Mean, %	10.769	10.769	10.774	10.776	10.769	10.774
St.Dev.	0.440	0.440	0.445	0.446	0.440	0.445
PLS factors	4	4	3	2	3	2
Slope	0.798928	0.794752	0.820999	0.665838	0.795888	0.647122
R^2 validation	0.799857	0.806851	0.816209	0.775217	0.806305	0.731694
RMSEP, %	0.196416	0.192949	0.190288	0.218102	0.193178	0.234211
SEP, %	0.196768	0.193390	0.190751	0.218611	0.193639	0.234616
Bias	0.007621	0.004451	0.002697	0.004402	0.003482	0.009397
RPD	2.236	2.275	2.333	2.040	2.272	1.897
Starch content as-is, complete dataset						
N	200	200	200	200	200	200
Mean, %	55.481	55.481	55.481	55.481	55.481	55.481
St.Dev.	2.096	2.096	2.096	2.096	2.096	2.096
PLS factors	5	3	3	3	4	2
Slope	0.320555	0.269179	0.518375	0.386237	0.521119	0.360265
R^2 validation	0.246234	0.240054	0.429895	0.328248	0.425700	0.318174
RMSEP, %	1.843405	1.827410	1.604018	1.727998	1.613994	1.734902
SEP, %	1.846496	1.831305	1.607559	1.730826	1.617601	1.737661
Bias	0.075102	0.050176	–0.039333	0.072111	–0.037814	0.074271
RPD	1.135	1.145	1.304	1.211	1.296	1.206
Starch content as-is, without outliers						
N	197	199	199	198	198	198
Mean, %	55.494	55.458	55.458	55.481	55.426	55.475
St.Dev.	2.006	2.075	2.075	2.106	2.029	2.105
PLS factors	5	5	3	3	4	4
Slope	0.370518	0.373981	0.524542	0.406056	0.535658	0.420821
R^2 validation	0.331003	0.340937	0.437978	0.361882	0.460178	0.371229
RMSEP, %	1.644813	1.684634	1.575651	1.690889	1.503929	1.675333
SEP, %	1.646675	1.688808	1.579255	1.689678	1.507491	1.678236
Bias	0.087377	0.015892	–0.034093	0.136058	–0.027421	–0.067011
RPD	1.218	1.229	1.314	1.246	1.346	1.254

(continued)

Table 4.11.1.2. (continued)

Statistics	Spectral data pre-treatment					
	Raw spectra	MSC	MSC + 1st Der.	MSC + 2nd Der.	1st Der.	2nd Der.
Starch content DWB, complete dataset						
N	200	200	200	200	200	200
Mean, %	62.181	62.181	62.181	62.181	62.181	62.181
St.Dev.	2.389	2.389	2.389	2.389	2.389	2.389
PLS factors	12	11	4	3	4	2
Slope	0.603839	0.561134	0.501566	0.345039	0.464712	0.333582
R^2 validation	0.450669	0.420679	0.452683	0.301494	0.418010	0.329888
RMSEP, %	1.848778	1.888259	1.771836	2.003914	1.82648	1.953780
SEP, %	1.853036	1.890897	1.776275	2.006031	1.831056	1.956108
Bias	–0.037528	–0.088942	–0.005243	–0.107858	–0.005028	–0.100137
RPD	1.289	1.263	1.345	1.191	1.305	1.221
Starch content DWB, without outliers						
N	197	197	199	198	199	198
Mean, %	62.196	62.196	62.145	62.163	62.145	62.171
St.Dev.	2.291	2.291	2.340	2.391	2.340	2.399
PLS factors	10	5	4	2	4	3
Slope	0.529386	0.389213	0.485017	0.305406	0.453429	0.376548
R^2 validation	0.420039	0.349335	0.434549	0.328036	0.408007	0.379313
RMSEP, %	1.782617	1.850577	1.764002	1.960217	1.803287	1.897941
SEP, %	1.787129	1.854574	1.768434	1.962349	1.807828	1.890162
Bias	0.010344	–0.051433	–0.007808	–0.105299	–0.004718	–0.217964
RPD	1.282	1.235	1.323	1.218	1.294	1.269

4.11.2. Calibration results for the 2007 season

Results of the 2007 season calibrations are presented in Table 4.11.2.1 for the whole kernels and in Table 4.11.2.2 for the milled triticale grain samples. In the 2007 season, dataset was increased to 220 samples from elite trials which were assessed for total starch and moisture content. The total starch content (‘as-is’) ranged between 51.78–62.99% (mean = 57.176%, SD = 2.231) and moisture content between 9.6–12.02% (mean = 10.738%, SD = 0.458). The PLS1 models based on whole grain spectra performed better than in the 2006 season (see Tables 4.11.2.1 and 4.11.1.1). For moisture content the best RPD was 1.691 units for the complete dataset with the best squared coefficient of determination $R^2 = 0.657$, SEP = 0.271% achieved without any pre-treatment (on raw spectra) with 11 PLS factors. The dataset cleaned from outliers resulted in the best RPD = 1.665 with $R^2 = 0.646$, SEP = 0.267% achieved

with the MSC data pre-treatment. For starch content ('as-is') the best RPD's were 1.646 (complete dataset, SEP = 1.356%) and 1.624 (without outliers, SEP = 1.38%), with R^2 *ca*. 0.634 achieved on raw spectra with 10 PLS factors. Results for starch content (DWB) were slightly worse with the best RPD for complete dataset of 1.532 units and $R^2 = 0.584$ achieved on raw spectra. For dataset cleaned from outliers the best pre-treatment was the 1st derivative, which resulted in RPD = 1.561, $R^2 = 0.596$.

Table 4.11.2.1. Cross-validation prediction results of the PLS1 calibration models with different data pre-treatments for starch and moisture contents in triticale whole kernels, the 2007 season

Statistics	Spectral data pre-treatment					
	Raw spectra	MSC	MSC + 1st Der.	MSC + 2nd Der.	1st Der.	2nd Der.
Moisture content, complete dataset						
N	220	220	220	220	220	220
Mean, %	10.738	10.738	10.738	10.738	10.738	10.738
St.Dev.	0.458	0.458	0.458	0.458	0.458	0.458
PLS factors	11	8	4	3	3	3
Slope	0.726428	0.709227	0.711252	0.482180	0.640610	0.477890
R^2 validation	0.656805	0.648112	0.644367	0.529159	0.637704	0.511763
RMSEP, %	0.270414	0.273005	0.275023	0.314689	0.275335	0.319729
SEP, %	0.270906	0.273627	0.275503	0.315341	0.275402	0.320434
Bias	0.008199	0.000593	0.008987	0.006400	0.017533	0.003893
RPD	1.691	1.674	1.662	1.452	1.663	1.429
Moisture content, without outliers						
N	218	218	219	219	219	218
Mean, %	10.726	10.726	10.732	10.733	10.732	10.728
St.Dev.	0.444	0.444	0.450	0.454	0.452	0.448
PLS factors	10	8	4	3	3	3
Slope	0.733034	0.710567	0.711004	0.523598	0.635276	0.487169
R^2 validation	0.644111	0.645757	0.640104	0.567719	0.615014	0.528282
RMSEP, %	0.268836	0.266057	0.272578	0.266078	0.280124	0.307984
SEP, %	0.269393	0.266665	0.273041	0.299745	0.280391	0.308686
Bias	0.005758	0.001424	0.009366	0.003233	0.014469	–0.002134
RPD	1.648	1.665	1.648	1.515	1.612	1.451

(continued)

Table 4.11.2.1. (continued)

Statistics	Spectral data pre-treatment					
	Raw spectra	MSC	MSC + 1st Der.	MSC + 2nd Der.	1st Der.	2nd Der.
Starch content as-is, complete dataset						
N	220	220	220	220	220	220
Mean, %	57.176	57.176	57.176	57.176	57.176	57.176
St.Dev.	2.231	2.231	2.231	2.231	2.231	2.231
PLS factors	10	8	4	2	4	1
Slope	0.686247	0.662004	0.616263	0.489555	0.617373	0.475803
R^2 validation	0.634755	0.610506	0.573220	0.463286	0.576770	0.469581
RMSEP, %	1.353276	1.396679	1.462271	1.634218	1.455490	1.620955
SEP, %	1.355726	1.399809	1.462683	1.636376	1.455993	1.624624
Bias	0.041444	0.012501	0.092306	0.071519	0.090399	0.009531
RPD	1.646	1.594	1.525	1.363	1.532	1.373
Starch content as-is, without outliers						
N	216	218	219	219	219	219
Mean, %	57.172	57.174	57.182	57.159	57.169	57.159
St.Dev.	2.241	2.175	2.234	2.221	2.233	2.221
PLS factors	10	7	3	3	4	2
Slope	0.723472	0.647927	0.596964	0.491744	0.621183	0.512013
R^2 validation	0.633487	0.602178	0.581313	0.486310	0.594584	0.486024
RMSEP, %	1.381201	1.375388	1.444773	1.588137	1.422067	1.590814
SEP, %	1.380349	1.377873	1.446220	1.591645	1.423808	1.594144
Bias	0.105706	–0.043205	0.073260	0.020309	0.065574	0.031590
RPD	1.624	1.579	1.545	1.395	1.568	1.393
Starch content DWB, complete dataset						
N	220	220	220	220	220	220
Mean, %	64.053	64.053	64.053	64.053	64.053	64.053
St.Dev.	2.439	2.439	2.439	2.439	2.439	2.439
PLS factors	10	6	3	2	3	1
Slope	0.659162	0.590159	0.601878	0.458682	0.601351	0.444899
R^2 validation	0.583771	0.578334	0.567477	0.426460	0.552279	0.444981
RMSEP, %	1.588563	1.580773	1.604437	1.848069	1.636421	1.813020
SEP, %	1.591888	1.584286	1.607955	1.851085	1.639957	1.817100
Bias	0.030720	–0.017007	–0.021202	0.066483	–0.025251	0.014117
RPD	1.532	1.539	1.517	1.318	1.487	1.342
Starch content DWB, without outliers						
N	216	218	219	219	219	219
Mean, %	64.020	64.053	64.081	64.037	64.045	64.037
St.Dev.	2.445	2.376	2.408	2.434	2.442	2.434
PLS factors	9	6	3	2	4	1
Slope	0.677488	0.572815	0.589056	0.456489	0.657223	0.452904
R^2 validation	0.586981	0.564691	0.559555	0.429163	0.596052	0.449841
RMSEP, %	1.594470	1.564491	1.597479	1.838019	1.561493	1.801061
SEP, %	1.597642	1.568068	1.601137	1.841507	1.564015	1.805130
Bias	–0.041114	–0.008639	–0.002504	0.051491	–0.057333	0.014277
RPD	1.530	1.515	1.504	1.322	1.561	1.348

Calibrations developed from milled grain spectra gave much better results for moisture content compared to the whole kernels samples (Table 4.11.2.2). The best calibration for moisture content on milled grain of the 2007 season was slightly better compared to such developed in the 2006 season: the MSC + 1st derivative pre-treatment without outliers had RPD = 2.526 units with R^2 = 0.843, SEP = 0.182% and 2 PLS factors.

Calibrations for starch content 'as-is' done on complete dataset gave the best results with the 1st derivative pre-treatment (RPD = 1.743, R^2 = 0.671, SEP = 1.28%), and calibration with the same pre-treatment cleaned from outliers had RPD = 1.741, R^2 = 0.673 with SEP = 1.277% and 3 PLS factors. Calibrations for starch content on DWB gave slightly worse results (see Table 4.11.2.2).

The 2007 season calibrations had the MSC + 1st derivative and the 1st derivative alone as the best spectral data pre-treatments for milled grain samples, and for whole kernels the MSC alone in many cases was the best pre-treatment. General quality of calibrations had improved in the 2007 season, with moisture calibrations being possible to use for rough screening of samples, but calibrations for starch content still having low predictive power (WILLIAMS, 2001).

Table 4.11.2.2. Cross-validation prediction results of the PLS1 calibration models with different data pre-treatments for starch and moisture contents in triticale milled grain, the 2007 season

Statistics	Spectral data pre-treatment					
	Raw spectra	MSC	MSC + 1st Der.	MSC + 2nd Der.	1st Der.	2nd Der.
Moisture content, complete dataset						
N	220	220	220	220	220	220
Mean, %	10.738	10.738	10.738	10.738	10.738	10.738
St.Dev.	0.458	0.458	0.458	0.458	0.458	0.458
PLS factors	7	3	2	3	3	3
Slope	0.847571	0.820412	0.790974	0.657715	0.809523	0.631532
R^2 validation	0.807455	0.788081	0.803709	0.742517	0.799229	0.734420
RMSEP, %	0.201545	0.210929	0.202359	0.235987	0.204641	0.241639
SEP, %	0.201804	0.211285	0.202816	0.236495	0.205079	0.242105
Bias	–0.008992	–0.007242	0.001347	–0.003766	–0.003420	–0.006420
RPD	2.270	2.168	2.258	1.937	2.233	1.892
Moisture content, without outliers						
N	217	217	217	219	219	219
Mean, %	10.736	10.736	10.741	10.739	10.739	10.739
St.Dev.	0.455	0.456	0.460	0.458	0.458	0.458
PLS factors	4	3	2	3	3	3
Slope	0.802662	0.848940	0.834362	0.682477	0.822479	0.663912
R^2 validation	0.793019	0.798398	0.843161	0.768002	0.811927	0.762368
RMSEP, %	0.206559	0.205852	0.181671	0.224508	0.198151	0.228708
SEP, %	0.207035	0.206322	0.182089	0.224982	0.198605	0.229055
Bias	0.000875	–0.001566	0.000885	–0.004249	0.000313	–0.008985
RPD	2.198	2.210	2.526	2.036	2.306	2.000
Starch content as-is, complete dataset						
N	220	220	220	220	220	220
Mean, %	57.176	57.176	57.176	57.176	57.176	57.176
St.Dev.	2.231	2.231	2.231	2.231	2.231	2.231
PLS factors	4	2	3	3	3	2
Slope	0.643530	0.618102	0.690465	0.598622	0.692387	0.624278
R^2 validation	0.628985	0.624007	0.661148	0.619567	0.671264	0.629455
RMSEP, %	1.356363	1.364784	1.298149	1.373946	1.277307	1.355028
SEP, %	1.359259	1.367829	1.300907	1.377058	1.280182	1.357862
Bias	0.023051	0.013565	0.022860	–0.007668	0.009877	0.026296
RPD	1.641	1.631	1.715	1.620	1.743	1.643
Starch content as-is, without outliers						
N	217	218	218	219	219	218
Mean, %	57.192	57.134	57.134	57.161	57.161	57.134
St.Dev.	2.165	2.193	2.193	2.223	2.223	2.193
PLS factors	4	2	3	3	3	1
Slope	0.653720	0.642812	0.712381	0.644055	0.712623	0.601383
R^2 validation	0.627554	0.624980	0.668848	0.651343	0.672592	0.655570
RMSEP, %	1.320520	1.340623	1.264306	1.309955	1.273875	1.292373
SEP, %	1.323364	1.343702	1.267202	1.312909	1.276743	1.295202
Bias	0.023455	0.004342	0.005953	0.011144	–0.011296	0.019403
RPD	1.636	1.632	1.731	1.693	1.741	1.693

(continued)

Table 4.11.2.2. (continued)

Statistics	Spectral data pre-treatment					
	Raw spectra	MSC	MSC + 1st Der.	MSC + 2nd Der.	1st Der.	2nd Der.
Starch content DWB, complete dataset						
N	220	220	220	220	220	220
Mean, %	64.053	64.053	64.053	64.053	64.053	64.053
St.Dev.	2.439	2.439	2.439	2.439	2.439	2.439
PLS factors	4	2	2	2	3	1
Slope	0.604491	0.600282	0.632619	0.607925	0.683321	0.599700
R^2 validation	0.587729	0.606833	0.609325	0.593735	0.635797	0.627936
RMSEP, %	1.563727	1.526127	1.523445	1.554896	1.475990	1.487206
SEP, %	1.566995	1.529510	1.526255	1.555289	1.479123	1.490298
Bias	0.030490	0.017174	0.044954	0.098862	0.026209	0.029850
RPD	1.556	1.595	1.598	1.568	1.649	1.637
Starch content DWB, without outliers						
N	217	218	218	219	218	218
Mean, %	64.074	64.008	64.008	64.036	64.008	64.008
St.Dev.	2.361	2.400	2.400	2.432	2.400	2.400
PLS factors	4	3	3	2	3	1
Slope	0.619151	0.638640	0.728438	0.653593	0.713725	0.625109
R^2 validation	0.598471	0.628768	0.662646	0.635109	0.667746	0.655329
RMSEP, %	1.494026	1.459672	1.406457	1.470474	1.387470	1.409109
SEP, %	1.497478	1.462790	1.407587	1.470417	1.390156	1.412040
Bias	–0.002774	0.026507	0.076856	0.100204	0.037453	0.029616
RPD	1.577	1.641	1.705	1.654	1.726	1.700

4.11.3. Calibration results for the combined 2006 and 2007 season's datasets

Datasets from the 2006 and 2007 seasons were combined in order to check hypothesis that larger dataset with more data points may result in development of more robust, better quality calibrations. The combined dataset consisted of 420 samples with moisture content range between 9.51–12.02% (mean = 10.753%, SD = 0.455), starch content ('as-is') between 49.46–62.99% (mean = 56.369%, SD = 2.325) and starch content (DWB) between 55.71–70.31% (mean = 63.161%, SD = 2.588). Development of calibrations was done for each trait only for those pre-treatments or raw data which showed good results in one of calculations for individual 2006 or 2007 season's datasets discussed in previous sections.

For the whole kernels samples (Table 4.11.3.1) the best calibration for moisture content was achieved for the dataset cleaned from outliers with the MSC + 1st derivative data pre-treatment, which was characterised by the RPD = 1.327, R^2 = 0.437 and SEP = 0.338%. The result of the calibration performance was worse than for any of previously developed calibrations from the 2006 or 2007 season's datasets taken individually.

For the starch content ('as-is') the best calibration for the combined dataset was the MSC + 1st derivative pre-treatment without outliers (RPD = 1.385, R^2 = 0.483, SEP = 1.665% with 5 PLS factors). Calibration for starch content (DWB) had slightly worse characteristics (RPD = 1.362, R^2 = 0.473, SEP = 1.879%). Performance of calibrations for starch content developed from the combined 2006–2007 dataset was better than that of the 2006 but worse than that of the 2007 season's datasets, assessed individually. Therefore, the results did not confirm the abovestated hypothesis.

Table 4.11.3.1. Cross-validation prediction results of the PLS1 calibration models with different data pre-treatments for starch and moisture contents in triticale whole kernels, combined 2006-2007 seasons dataset

Statistics	Spectral data pre-treatment					
	Raw spectra	MSC	MSC + 1st Der.	MSC + 2nd Der.	1st Der.	2nd Der.
Moisture content, complete dataset						
N	420	420	420	420	.	.
Mean, %	10.753	10.753	10.753	10.753	.	.
St.Dev.	0.455	0.455	0.455	0.455	.	.
PLS factors	12	10	6	3	.	.
Slope	0.480603	0.451808	0.451670	0.366217	.	.
R^2 validation	0.415333	0.400208	0.385737	0.298879	.	.
RMSEP, %	0.350904	0.354007	0.360501	0.384631	.	.
SEP, %	0.350947	0.354329	0.359862	0.385084	.	.
Bias	0.016217	0.008429	0.027723	0.002026	.	.
RPD	1.296	1.284	1.264	1.182	.	.
Moisture content, without outliers						
N	417	418	417	419	.	.
Mean, %	10.751	10.753	10.752	10.756	.	.
St.Dev.	0.446	0.449	0.449	0.451	.	.
PLS factors	12	10	6	3	.	.
Slope	0.492913	0.463252	0.487860	0.372526	.	.
R^2 validation	0.422208	0.408467	0.436543	0.306784	.	.
RMSEP, %	0.342326	0.347143	0.338433	0.379234	.	.
SEP, %	0.342405	0.347497	0.338474	0.379683	.	.
Bias	0.015074	0.006567	0.015704	0.001719	.	.
RPD	1.303	1.292	1.327	1.188	.	.
Starch content as-is, complete dataset						
N	420	420	420	.	420	.
Mean, %	56.369	56.369	56.369	.	56.369	.
St.Dev.	2.325	2.325	2.325	.	2.325	.
PLS factors	10	7	4	.	3	.
Slope	0.504276	0.438358	0.470188	.	0.441538	.
R^2 validation	0.457287	0.403878	0.400851	.	0.401552	.
RMSEP, %	1.718503	1.797810	1.816991	.	1.802885	.
SEP, %	1.720303	1.799423	1.817469	.	1.804468	.
Bias	0.029334	0.043691	0.078271	.	0.045221	.
RPD	1.352	1.292	1.279	.	1.288	.
Starch content as-is, without outliers						
N	417	416	418	.	418	.
Mean, %	56.343	56.359	56.347	.	56.347	.
St.Dev.	2.306	2.286	2.307	.	2.307	.
PLS factors	9	7	5	.	4	.
Slope	0.491329	0.469756	0.525037	.	0.503370	.
R^2 validation	0.467213	0.450293	0.482722	.	0.467956	.
RMSEP, %	1.682907	1.693870	1.663510	.	1.685126	.
SEP, %	1.684921	1.695876	1.665317	.	1.687129	.
Bias	–0.005120	0.010790	0.024907	.	0.007416	.
RPD	1.369	1.348	1.385	.	1.367	.

(continued)

Table 4.11.3.1. (continued)

Statistics	Spectral data pre-treatment					
	Raw spectra	MSC	MSC + 1st Der.	MSC + 2nd Der.	1st Der.	2nd Der.
Starch content DWB, complete dataset						
N	420	420	420	.	420	.
Mean, %	63.161	63.161	63.161	.	63.161	.
St.Dev.	2.588	2.588	2.588	.	2.588	.
PLS factors	10	7	4	.	4	.
Slope	0.486097	0.427088	0.411526	.	0.436010	.
R^2 validation	0.414369	0.383985	0.352220	.	0.373893	.
RMSEP, %	1.999115	2.037172	2.096937	.	2.062145	.
SEP, %	2.001338	2.039142	2.098884	.	2.064558	.
Bias	0.025402	0.043270	0.048169	.	0.013803	.
RPD	1.293	1.269	1.233	.	1.254	.
Starch content DWB, without outliers						
N	418	417	417	.	418	.
Mean, %	63.137	63.165	63.124	.	63.137	.
St.Dev.	2.568	2.557	2.558	.	2.569	.
PLS factors	10	7	5	.	4	.
Slope	0.515966	0.452348	0.549754	.	0.494531	.
R^2 validation	0.449813	0.426128	0.473205	.	0.452500	.
RMSEP, %	1.919290	1.937780	1.876309	.	1.905801	.
SEP, %	1.921580	1.940108	1.878563	.	1.907856	.
Bias	0.005906	–0.000499	0.000864	.	0.029459	.
RPD	1.336	1.318	1.362	.	1.347	.

For the milled grain samples (Table 4.11.3.2) the best calibration for moisture content was developed from a dataset cleaned from outliers with the MSC + 1st derivative pre-treatment and the RPD = 2.244, R^2 = 0.802, SEP = 0.2%. Performance of the calibration was worse than any of the developed from the 2006 or 2007 season's datasets taken individually (same as for whole kernels).

For starch content ('as-is') the best calibration for the combined dataset was the 1st derivative pre-treatment without outliers (RPD = 1.527, R^2 = 0.572, SEP = 1.52% with 5 PLS factors). Calibration for starch content (DWB) had a bit worse characteristics (RPD = 1.496, R^2 = 0.56, SEP = 1.71%). Performance of calibrations for starch content developed from the combined dataset was in between of the best relevant calibrations of individual season's datasets (better than in the 2006 but worse than in the 2007 season), similar to the developed from whole kernels data.

Table 4.11.3.2. Cross-validation prediction results of the PLS1 calibration models with different data pre-treatments for starch and moisture contents in triticale milled grain, combined 2006-2007 seasons dataset

Statistics	Spectral data pre-treatment					
	Raw spectra	MSC	MSC + 1st Der.	MSC + 2nd Der.	1st Der.	2nd Der.
Moisture content, complete dataset						
N	420	420	420	.	420	.
Mean, %	10.753	10.753	10.753	.	10.753	.
St.Dev.	0.455	0.455	0.455	.	0.455	.
PLS factors	5	6	4	.	5	.
Slope	0.774484	0.772469	0.783285	.	0.780094	.
R^2 validation	0.767465	0.763507	0.761494	.	0.754238	.
RMSEP, %	0.219178	0.221054	0.222300	.	0.226002	.
SEP, %	0.219439	0.221318	0.222499	.	0.225969	.
Bias	–0.000073	–0.000173	0.005437	.	0.011682	.
RPD	2.073	2.056	2.045	.	2.014	.
Moisture content, without outliers						
N	418	415	417	.	417	.
Mean, %	10.757	10.755	10.754	.	10.754	.
St.Dev.	0.451	0.452	0.449	.	0.449	.
PLS factors	5	5	4	.	5	.
Slope	0.797160	0.794376	0.811367	.	0.808412	.
R^2 validation	0.789056	0.788675	0.801657	.	0.782942	.
RMSEP, %	0.206914	0.207515	0.199857	.	0.209528	.
SEP, %	0.207159	0.207759	0.200077	.	0.209760	.
Bias	–0.001119	0.001653	–0.002830	.	–0.002811	.
RPD	2.177	2.176	2.244	.	2.141	.
Starch content as-is, complete dataset						
N	420	420	420	.	420	.
Mean, %	56.369	56.369	56.369	.	56.369	.
St.Dev.	2.325	2.325	2.325	.	2.325	.
PLS factors	7	7	4	.	4	.
Slope	0.515445	0.540650	0.574513	.	0.572889	.
R^2 validation	0.488668	0.496191	0.542017	.	0.552348	.
RMSEP, %	1.662849	1.654957	1.574907	.	1.554964	.
SEP, %	1.664830	1.656683	1.576677	.	1.556812	.
Bias	–0.002417	–0.028644	–0.018428	.	–0.004536	.
RPD	1.397	1.403	1.475	.	1.493	.
Starch content as-is, without outliers						
N	418	419	419	.	419	.
Mean, %	56.340	56.360	56.360	.	56.360	.
St.Dev.	2.291	2.320	2.320	.	2.320	.
PLS factors	7	7	5	.	5	.
Slope	0.518204	0.548291	0.601100	.	0.596322	.
R^2 validation	0.493551	0.527382	0.571055	.	0.572147	.
RMSEP, %	0.630768	1.595162	1.521850	.	1.519237	.
SEP, %	1.632719	1.596646	1.522552	.	1.519631	.
Bias	–0.003236	–0.036679	–0.058256	.	–0.065674	.
RPD	1.403	1.453	1.524	.	1.527	.

(continued)

Table 4.11.3.2. (continued)

Statistics	Spectral data pre-treatment					
	Raw spectra	MSC	MSC + 1st Der.	MSC + 2nd Der.	1st Der.	2nd Der.
Starch content DWB, complete dataset						
N	420	420	420	.	420	420
Mean, %	63.161	63.161	63.161	.	63.161	63.161
St.Dev.	2.588	2.588	2.588	.	2.588	2.588
PLS factors	10	8	5	.	3	2
Slope	0.574203	0.546718	0.610103	.	0.563535	0.511133
R^2 validation	0.520031	0.514567	0.555567	.	0.518017	0.468732
RMSEP, %	1.801444	1.804856	1.733570	.	1.802226	1.891176
SEP, %	1.803422	1.806818	1.735624	.	1.804112	1.893093
Bias	0.024778	–0.026250	–0.006634	.	0.030817	0.035768
RPD	1.435	1.432	1.491	.	1.435	1.367
Starch content DWB, without outliers						
N	416	417	417	.	417	.
Mean, %	63.119	63.127	63.124	.	63.124	.
St.Dev.	2.551	2.563	2.558	.	2.558	.
PLS factors	9	8	5	.	3	.
Slope	0.569739	0.558745	0.619074	.	0.578470	.
R^2 validation	0.533693	0.520196	0.559507	.	0.537254	.
RMSEP, %	1.744674	1.779128	1.708189	.	1.744206	.
SEP, %	1.746587	1.780828	1.710148	.	1.746264	.
Bias	0.025584	–0.039439	–0.017797	.	0.011505	.
RPD	1.461	1.439	1.496	.	1.465	.

One of the aims of this study was to develop NIR spectroscopy prediction models which could be used as a surrogate rapid quality assessment tool for starch and moisture contents in triticale grain. This was intended to achieve by applying different spectral data pre-treatments to the datasets, and by comparing performances of the calibration models, built from these spectra and reference test data. The study demonstrated that NIR spectroscopy calibration models have the potential to be used in the triticale breeding programme as a screening technique instead of conventional analytical methods for the rapid prediction of the traits involved. The moisture content calibration model gave acceptable prediction results for the purpose of screening. However, it was not possible to accurately predict starch content in triticale grain and whole-ground flour using NIR spectroscopy calibration models. The best suitable data pre-treatments in most cases were the MSC and the 1st derivative, as well as their

combination. Pooling of datasets from the 2006 and 2007 seasons did not result in more robust prediction models, with the 2007 season results remained the best. Improvement of the calibration models is possible if wider genetic pool of newer breeding material created in the triticale breeding programme could be involved, giving wider range of values for the traits investigated. Exploration of the possibility of the development of NIRS prediction models for direct assessment of ethanol output and ethanol yield from whole grain or milled samples would be recommended.

Chapter 5: Conclusions and recommendations

The area under investigation is located in a zone with a Mediterranean-like climate and has an insufficiently wet climatic regimen with two sub-environments, namely, Swartland and Overberg regions, with different soils and weather patterns resulting in their different yield potential. The 2006 and 2007 seasons were characterised by higher (compared to long-term data) amounts of precipitation, with generally favourable weather conditions, especially in the 2007 season. Spatial data analysis performed for individual trials helped to eliminate spatial differences of plots within and between blocks. It could be recommended that trials in Tygerhoek and Mariendahl locations be relocated to other testing locations, or resources allocated for these trial locations be reallocated to other research activities, as the Tygerhoek location is redundant, and Mariendahl does not represent the target region.

Use of standard AACC International Method 44-15.02 for moisture determination is recommended as opposed to the moisture scale method if more accurate results are to be achieved. It could be recommended that sub-samples should be taken from each replication of a trial and mixed into a composite sample prior to analyses. Better (standard) method of protein determination is recommended, which would give more realistic results for the trait. Additionally, it would be recommended to use standard 'normal gravity' (20–24°Plato) levels of solids concentration in small-scale fermentations, as opposed to 17.2°Plato used in this research.

The AAQ values were much lower than expected, which can be partially explained by the fact that a conventional step of jet cooking was not performed. Another possible reason is more acidic pH (3.6 against recommended 5.0–5.8) of the mash during gelatinisation and fermentation stages, which could hinder endogenous enzymes performance.

NIRS calibrations developed from the 2006 season samples were weak in their predictive power. Best calibrations for moisture content were on whole kernels with RPD = 1.320, R^2 = 0.441, SEP = 0.333% and 10 PLS factors, and on milled grain with RPD = 2.333, R^2 = 0.816, SEP = 0.191% and 3 PLS factors. For starch content ('as-is') on whole kernels best calibration had RPD = 1.095, R^2 = 0.2; on milled grain had RPD = 1.346, R^2 = 0.46, SEP = 1.507% and 4 PLS factors.

In the 2007 season, NIRS calibration models based on whole grain spectra performed better than those developed from the 2006 season dataset. For moisture content best calibration had RPD = 1.691, R^2 = 0.657, SEP = 0.271% and 11 PLS factors. For starch content ('as-is') best model had RPD = 1.646, R^2 = 0.634, SEP = 1.356% with 10 PLS factors. Calibrations developed from milled grain showed better results. The best calibration for moisture content had RPD = 2.526, R^2 = 0.843, SEP = 0.182% and 2 PLS factors, and for starch content ('as-is') had RPD = 1.741, R^2 = 0.673, SEP = 1.277% and 3 PLS factors.

Pooling of 2006 and 2007 season datasets did not result in development of more robust, better quality calibrations as was anticipated. In some cases best resulting calibration performed even worse than any of the calibrations developed from individual 2006 or 2007 season dataset; in other cases its performance was better than for models developed from the 2006 season dataset but worse than that of the 2007 season dataset, irrespective of samples used (whole kernels or milled grain).

Parameters of calibrations (e.g. RPD) for moisture content from both seasons proved good enough for very rough screening of samples and their ranking; however, calibrations for starch content were characterised by very low predictive power and cannot be recommended for practical use. The best spectral data pre-treatments in most cases were MSC, 1st derivative and their combination for both whole kernels and

milled grain samples. Exclusion of outliers was beneficial for the development of more robust calibrations. NIR spectroscopy calibration models have demonstrated the potential for use in the triticale breeding programme as a screening technique instead of conventional analytical methods for the rapid prediction of the traits involved. Improvement of the calibration models is possible if a wider genetic pool of newer breeding material created in the triticale breeding programme could be involved, thus resulting in a wider range of values for the investigated traits. Application of more reliable reference testing methods would be also beneficial. It might be recommended that the possibility of a calibration development for such traits as ethanol output and ethanol yield be investigated, which are of the utmost interest in relation to the investigated subject.

The most important traits for this study were assessed, namely, grain yield ($kg.ha^{-1}$), total starch content (%) and starch yield ($kg.ha^{-1}$), ethanol output ($L.tonne^{-1}$) and ethanol yield ($L.ha^{-1}$), as well as disease resistance. During both 2006 and 2007 seasons, most traits under investigation were not normally distributed, thus non-parametric methods of analysis were employed.

In the 2006 season, starch yield was highly positively correlated with grain yield ($R^2 = 0.988$, $P < 0.001$). Both starch yield and grain yield were positively correlated with days to heading ($R^2 = 0.533$ and $R^2 = 0.556$, respectively; $P < 0.001$). Longer days to heading could be cautiously used as an indirect selection trait for higher grain and starch yields.

The 2007 season was characterised by a generally higher starch yield (2952–3142$kg.ha^{-1}$, 95%CI) compared to the 2006 season (2077–2315$kg.ha^{-1}$, 95%CI). Starch yield was strongly positively correlated with grain yield ($R^2 = 0.975$, $P < 0.001$).

The best lines for starch yield in the 2006 season were YB and CA for the Swartland region, and Y3, CA and Y2 for the Overberg. These lines could be recommended as initial material for the development of new high starch yielding cultivars for the respective regions.

The best lines in terms of starch yield in the 2007 season for the Swartland region were H3, G2 and EA, and for the Overberg region G2 and D2. They can be recommended as initial material for the breeding programme as donors of high starch yield in the respective regions.

Test weight demonstrated weak positive correlation with ethanol yield ($R^2 = 0.238$, $P < 0.01$) and grain yield ($R^2 = 0.279$, $P < 0.001$). The best lines for the test weight were EB and Y2. However, they were the ones on the lower end for ethanol yield, as well as for ethanol output. Taking the above into consideration, test weight cannot be used as a reliable predictor of genotypes performance for ethanol output and ethanol yield or used for indirect selection for these traits. Assessment of other traits, such as kernel length to width ratio, thousand-kernel weight, number of grains in ear, residue viscosity etc. could be recommended.

Mean ethanol output ranged between 466–477L.tonne^{-1} at the 95%CI. It can be concluded that selection for higher ethanol output would not have much success due to low variation of the trait among the investigated lines. Ethanol output is demonstrated to be more dependent on starch and other polysaccharides accessibility to enzymatic digestion than on the total starch content as such. The best lines for ethanol output in the 2007 season were G2, D3 and H2 for the Swartland region, and D3, G2 and D1 for the Overberg region, on the contrary to the best in terms of total starch content lines H6, G1, H3 and D4 (across all locations). Thus, the total starch

content taken as a possible indirect predictor of the ethanol output without consideration of the actual ethanol output can lead to erroneous conclusions.

Correlation pattern of the ethanol yield with other traits closely followed the correlation pattern of starch yield, both traits being highly positively correlated ($R^2 = 0.91$, $P < 0.001$). Correlation of total starch content and protein content with ethanol yield did not confirm the hypothesis that they are highly correlated, thus these traits cannot serve as reliable predictors for the ethanol yield. On the other hand, relative moisture content has demonstrated better correlations with starch yield, ethanol output and ethanol yield.

The best triticale lines under investigation showed their potential from a biological point of view to be a suitable crop for ethanol production in the Western Cape, with the achieved ethanol yield ranging between 2446–2625L.ha^{-1} at the 95%CI. It had higher variability compared to ethanol output, thus successful selection for this trait is anticipated. For the Swartland region the best genotypes for ethanol yield were D1, H1 and D2, and for the Overberg H1 and G2. It might be recommended as promising breeding combinations for ethanol yield D1 / H1, CD / D1, CD / D2 and CD / H1 for the Swartland and H1 / G2 for the Overberg, as well as D2 / H1 and D2 / G2 for both sub-environments. The 23 best lines were selected from the elite and senior blocks, and then used for the establishment of marker-assisted recurrent selection pre-breeding block, which would lead to a high-ethanol yielding nursery.

References

AACC, 2000. Approved Methods of American Association of Cereal Chemists. 10th ed. AACC, St. Paul, MN, USA, 1200 pp.

ABDEL-AAL, E.-S.M., HUCL, P., CHIBBAR, R.N., HAN, H.L. & DEMEKE, T., 2002. Physicochemical and structural characteristics of flours and starches from waxy and nonwaxy wheats. *Cereal Chem*. 79: 458-464.

ABDI, H., 2007. Partial Least Square Regression. In: Neil Salkind (Ed.), *Encyclopedia of Measurement and Statistics*. Thousand Oaks (CA): Sage, pp. 1-13. Retrieved November 10, 2008, from http://www.utdallas.edu/~herve/Abdi-PLSR2007-pretty.pdf

ABE, H., KUSAMA, T., KAWANO, S., & IWAMOTO, M., 1996. Non-destructive determination of protein content in a single kernel of wheat and soybean by near-infrared spectroscopy. In: A.M.C. Davies, P. Williams (Eds.), *Near Infrared Spectroscopy: Future Waves*. NIR Publications: Chichester, UK, pp. 457-461.

ABECASSIS, J., 1993. Nouvelles possibilites d'apprecier la valeur meuniere et la valeur semouliere des bles. *Industrie des Cereales* 81: 25-37.

AKERBERG, A., LILJEBERG, H. & BJORCK, I., 1998. Effects of amylose/amylopectin ratio and baking conditions on resistant starch formation and glycaemic indices. *J. Cereal Sci.* 28: 71-80.

ALDHOUS, P., 2008. Genes for greens. *New Scientist* 197(2637): 28-31.

ALI, J., 2002. Functional starches. *Chem. Ind*. 6: 18-20.

ANDE, B., PIEPER, H.-J. & SENN, T., 1998. Fremdenzymfreie herstellung von glucosesirup durch direktverzuckerung aus autoamylolytisch hochaktivem triticale [Production of glucose syrup by direct saccharification from triticale with high autoamylolytic activity]. *Starch/Stärke* 50(11-12): 518-523.

ANDERSSON, A.A.M., ANDERSSON, R., AUTIO, K. & AMAN, P., 1999a. Chemical composition and microstructure of two naked waxy barleys. *J. Cereal Sci*. 30: 183-191.

ANDERSSON, L., FREDRIKSSON, H., OSCARSSON BERGH, M., ANDERSSON, R. & AMAN, P., 1999b. Characterisation of starch from inner and peripheral parts of normal and waxy barley kernels. *J. Cereal Sci*. 30: 165-171.

ANKRAH, N.O., CAMPBELL, G.L., TYLER, R.T., ROSSNAGEL, B.G. & SOKHANSANJ, S.R.T., 1999. Hydrothermal and β-glucanase effects on the nutritional and physical properties of starch in normal and waxy hull-less barley. *Anim. Feed Sci. Technol*. 81: 205-219.

ANNICCHIARICO, P., 2002. Genotype x environment interactions – challenges and opportunities for plant breeding and cultivar recommendations. *FAO plant production and protection paper*, 174. FAO, Rome, Italy. Retrieved November 4, 2007, from http://www.fao.org/docrep/005/Y4391E/y4391e00.HTM

ANONYMOUS, 1985. Corn Analysis: Waxy and Nonwaxy. *Analytical Methods of the Member Companies of the Corn Refiners Association, Inc*. Accepted 11-17-59, Revised 11-14-85, p. 1-3. Retrieved November 10, 2008, from http://www.corn.org/methods/B-64.pdf

ANONYMOUS, 1991. Corn Starch Analysis: Starch Identification (Microscopy). *Analytical Methods of the Member Companies of the Corn Refiners Association, Inc*. Accepted 6-20-91, p. 1-8. Retrieved November 10, 2008, from http://www.corn.org/methods/B-25.pdf

ANONYMOUS, 2000. Fertiliser Recommendations. 7th Edition. ISBN 0-11-243058-9. *Ministry of Agriculture Fisheries and Food*. The Stationary Office, London. Retrieved November 10, 2008, from http://www.defra.gov.uk/FARM/environment/land-manage/nutrient/fert/rb209/

ANONYMOUS, 2003. Driving to better ethanol hybrids. Trait characterization is pinpointing specific hybrids for the ethanol markets. *PGP magazine* Sep.-Oct. 2003: 4-5.

ANONYMOUS, 2006. An Investigation into the Feasibility of Establishing a Biofuels Industry in the Republic of South Africa. National Biofuels Study. *Final Report to the National Biofuels Task Team*, 6 October 2006, 116 pp.

ANONYMOUS, 2008. Agrofuels and the Myth of the Marginal Lands. *A briefing by The Gaia Foundation, Biofuelwatch, the African Biodiversity Network, Salva La Selva, Watch Indonesia and EcoNexus*. September 2008, pp. 1-8. Retrieved November 10, 2008, from http://www.africanbiodiversity.org/media/1221812708.pdf

AO, Z. & JANE, J.-I., 2007. Characterization and modeling of the A- and B-granule starches of wheat, triticale, and barley. *Carbohydr. Polym.* 67(1): 46-55.

ARAKI, E., MIURA, H. & SAWADA, S., 1999. Identification of genetic loci affecting amylose content and agronomic traits on chromosome 4A of wheat. *Theor. Appl. Genet*. 98: 977-984.

ARAKI, E., MIURA, H. & SAWADA, S., 2000. Differential effects of the null alleles at the three *Wx* loci on the starch properties of wheat. *Theor. Appl. Genet*. 100: 1113-1120.

ARCADIA BIOSCIENCES INC., 2007. Arcadia biosciences and Australian research organizations enter research and commercial license agreement for development of nitrogen use efficient wheat. Press release. *Arcadia Biosciences Inc*. Davis, California, USA, Adelaide, SA, Australia, and Canberra, ACT, Australia, October 10, 2007. Retrieved November 10, 2008, from http://www.arcadiabio.com/assets/media/71010-ACPFG-CSIRO.pdf

ASANO, T., KUNIEDA, N., OMURA, Y., IBE, H., KAWASAKI, T., TAKANO, M., SATO, M., FURUHASHI, H., MUJIN, T., TAKAIWA, F., WU, C.-Y., TADA, Y., SATOZAWA, T., SAKAMOTO, M., & SHIMADA, H., 2002. Rice SPK, a calmodulin-like domain protein kinase, is required for storage product accumulation during seed development: phosphorylation of sucrose synthase is a possible factor. *Plant Cell* 14: 619-628.

ASAOKA, M., OKUNO, K., SUGIMOLO, Y., KAWAKAMI, J. & FUWA, H., 1984. Effect of environmental temperature during development of rice plants on some properties of endosperm starch. *Starch/Stärke* 36: 189-193.

ASAOKA, M., OKUNO, K., SUGIMOTO, Y., OMURA, T., FUWA, H., 1986. Characterization of endosperm starch from high-amylose mutants of rice (*Oryza sativa* L.). *Starch/Stärke* 38: 114-117.

AUFHAMMER, W., 1998. Bioethanol yield of different varieties and variety mixtures of winter wheat and triticale. *BioBase European Energy Crops InterNetwork*. Retrieved November 16, 2007, from http://www.eeci.net/archive/biobase/B10381.html

AUFHAMMER, W., PIEPER, H.J., KASSER, J., SCHAFER, V., SENN, T. & KUBLER, E., 1996. Zur Eignung des Kornguts unterschiedlich stickstoffgedüngter Getreidebestände als Rohstoff für die Bioethanolproduktion (The suitability of grains from cereal crops with different N supply for bioethanol production). *Zeitschrift fur Acker und Pflanzenbau / Journal of Agronomy and Crop Science* 177: 185-196.

AUFHAMMER, W., PIEPER, H.J., KUBLER, E. & SCHAFER, V., 1994. Eignung des korngutes von weizen und triticale fur die bioethanolproducktion in abhangigkeit vom verarbeitungszeitpunkt nach der reife. *Bodenkultur* 45: 177-187.

AUFRÈRE, J., GRAVIOU, D., DEMARQUILLY, C., PEREZ, J.M. & ANDRIEU, J., 1996. Near infrared reflectance spectroscopy to predict energy value of compound feeds for swine and ruminants. *Anim. Feed Sci. Technol*. 62: 77-90.

BABA, T., YOSHII, M. & KAINUMA, K., 1987. Acceptor molecule of granulo-bound starch synthase from sweet-potato roots. *Starch/Stärke* 39(2): 52-56.

BADENHUIZEN, N.P., 1963. Formation and distribution of amylose and amylopectin in the starch granule. *Nature* 197: 464-467.

BADENHUIZEN, N.P., 1965. Occurrence and development of starch in plants. In: Whistler, R., BeMiller, J., Paschall, E. (Eds.), *Starch Chemistry and Technology, Vol 1*. Academic Press, New York, pp. 65-103.

BAE, J.M., GIROUX, M. & HANNAH, L., 1990. Cloning and characterization of the brittle-2 gene of maize. *Maydica* 35: 317-322.

BÅGA, M., GLAZE, S., MALLARD, C.S. & CHIBBAR, R.N., 1999a. A Starch Branching Enzyme Gene in Wheat Produces Alternatively Spliced Transcripts. *Plant Mol. Biol*. 40(6): 10-19.

BÅGA, M., REPELLIN, A., DEMEKE, T., CASWELL, K., LEUNG, N., ABDEL-AAL, E.S., HUCL, P. & CHIBBAR, R.N., 1999b. Wheat Starch Modification Through Biotechnology. *Starch/Stärke* 51(4): 111-116.

BAILEY, R., 2007. Bio-fuelling Poverty: Why the EU renewable-fuel target may be disastrous for poor people. *Oxfam Briefing Note*, November 2007. Retrieved February 16, 2008, from http://www.oxfam.org.uk/resources/policy/trade/downloads/bn_biofuels.pdf

BALAT, M., BALAT, H. & OZ, C., 2008. Progress in bioethanol processing. *Progress Energy Combust. Sci*. 34(5): 551-573.

BALDWIN, P.M., 2001. Starch granule-associated proteins and polypeptids: A review. *Starch/Stärke* 53: 475-503.

BALDWIN, P.M., ADLER, J., DAVIES, M.C. & MELIA, C.D., 1994. Holes in starch granules: confocal, SEM and light microscopy studies of starch granule structure. *Starch/Stärke* 46: 341-346.

BALDWIN, P.M., ADLER, M.C., DAVIES, M.C. & MELIA, C.D., 1998. High resolution imaging of starch granule surfaces by atomic force microscopy. *J. Cereal Sci*. 27: 255-265.

BALDWIN, P.M., DAVIES, M.C. & MELIA, C.D., 1997. Starch granule surface using low-voltage scanning electron microscopy and atomic force microscopy. *Int. J. Biol. Macromol*. 21: 103-107.

BALDWIN, P.M., MELIA, C.D. & DAVIES, M.C., 1997. The surface chemistry of starch granules studied by time-of-flight secondary ion mass spectrometry. *J. Cereal Sci.* 26: 329-346.

BALL, S.G., 1995. Recent views on the biosynthesis of the plant starch granule. *Trends in Glycoscience and Glycotechnology* 7: 405-415.

BALL, S., GUAN, H.-P., JAMES, M., MYERS, A., KELLING, P., MOUILLE, G., BULEON, A., COLONNA, P. & PREISS, J., 1996. From glycogen to amylopectin: A model for the biogenesis to the plant starch granule. *Cell* 86: 349-352.

BALL, S.G. & MORELL, M.K., 2003. From bacterial glycogen to starch: understanding the biogenesis of the plant starch granule. *Ann. Rev. Plant Biol.* 54: 207-233.

BALL, S., VAN DE WAL, M.H.B.J. & VISSER, R.G.F., 1998. Progress in understanding the biosynthesis of amylose. *Trends Plant Sci*. 3(3): 462-467.

BANKS, W., GREENWOOD, C.T. & MUIR, D.D., 1971. The characterization of starch and its components. Part 3. The technique of semimicro, differential, potentiometric titration, and the factors affecting it. *Starch/Stärke* 23: 118-127.

BAO, J.S., CAI, Y.Z. & CORKE, H., 2001. Prediction of rice starch quality parameters by near infrared reflectance spectroscopy. *J. Food Sci.* 66: 936-939.

BAO, J., SHEN, Y. & JIN, L., 2007. Determination of thermal and retrogradation properties of rice starch using near-infrared spectroscopy. *J. Cereal Sci.* 46(1): 75-81.

BAO, J.S., SHEN, S.Q., SUN, M. & CORKE, H., 2006. Analysis of genotypic diversity in the starch physicochemical properties of nonwaxy rice: apparent amylose content, pasting viscosity and gel texture. *Starch/Stärke* 58: 259-267.

BAO, J.S., WANG, Y.F. & SHEN, Y., 2007. Determination of apparent amylose content, pasting properties and gel texture of rice starch by near-infrared spectroscopy. *J. Sci. Food Agric.* 87(11): 2040-2048.

BAROJA-FERNÁNDEZ, E., ZANDUETA-CRIADO, A., RODRIGUEZ-LÓPEZ, M., AKAZAWA, T. & POZUETA-ROMERO, J., 2000. Distinct isoforms of ADPglucose pyrophosphatase and ADPglucose pyrophosphorylase occur in the suspension cultured cells of sycamore (*Acer pseudoplatanus* L.), *FEBS Lett*. 480: 277-282.

BARTON II, F.E. & WINDHAM, W.R., 1988. Determination of acid-detergent fiber and crude protein in forages by near-infrared reflectance spectroscopy: collaborative study. *J Assoc. Anal. Chem*. 71: 1162-1167.

BATEY, I.L. & CURTIN, B.M., 1996. Measurement of amylose/amylopectin ratio by high-performance liquid chromatography. *Starch/Stärke* 48: 338-344.

BAUM, B.R., 1971. Proposal to conserve the "generic name" *Triticale* Müntzing. *Taxon* 20: 644-645.

BAYE, T. & BECKER, H.C., 2004. Analyzing seed weight, fatty acid composition, oil, and protein contents in *Vernonia galamensis* germplasm by near-infrared reflectance spectroscopy. *J. Amer. Oil Chem. Soc.* 81: 641-645.

BAYE, T.M., PEARSON, T.C. & SETTLES, A.M., 2006. Development of a calibration to predict maize seed composition using single kernel near infrared spectroscopy. *J. Cereal Sci.* 43: 236-243.

BECHTEL, D.B. & WILSON, J.D., 2003. Amyloplast formation and starch granule development in hard red winter wheat. *Cereal Chem*. 80: 175-183.

BECHTEL, D.B., ZAYAS, I., KALEIKAU, L. & POMERANZ, Y., 1990. Size distribution of wheat starch granules during endosperm development. *Cereal Chem.* 67: 59-63.

BECKLES, D.M., SMITH, A.M. & REES, T., 2001. A cytosolic ADP-glucose pyrophosphorylase is a feature of graminaceous endosperms, but not of other starch-storing organs. *Plant Physiol*. 125: 818-827.

BEDNAR, G.E., PATIL, A.R., MURRAY, S.M., GRIESHOP, C.M., MERCHEN, N.R. & FAHEY JR., G.C., 2001. Starch and fiber fractions in selected food and feed ingredients affect their small intestinal digestibility and fermentability and their large bowel fermentability in vitro in a canine model. *J. Nutr*. 131: 276-286.

BEMILLER, J.N. & WHISTLER, R.L., 1996. Carbohydrates. In: O.R. Fennema (Ed.), *Food chemistry*, 3rd ed., New York: Marcel Dekker Inc., pp. 157-223.

BEN-GERA, I. & NORRIS, K.H., 1968. Determination of moisture content in soybeans by direct spectrophotometry. *J. Agric. Eng. Res.* 18(3): 125-132.

BERTOFT, E., 2004. On the nature of categories of chains in amylopectin and their connection to the super helix model. *Carbohydr. Polym.* 57: 211-224.

BERTOFT, E., 2007a. Composition of clusters and their arrangement in potato amylopectin. *Carbohydr. Polym.* 68: 433-446.

BERTOFT, E., 2007b. Composition of building blocks in clusters from potato amylopectin. *Carbohydr. Polym.* 70: 123-136.

BERTOLINI, A.C., SOUZA, E., NELSON, J.E. & HUBER, K.C., 2003. Composition and reactivity of A- and B-type starch granules of normal, partial waxy, and waxy wheat. *Cereal Chem.* 80(5): 544-549.

BETTGE, A.D., GIROUX, M.J. & MORRIS, C.F., 2000. Susceptibility of waxy starch granules to mechanical damage. *Cereal Chem.* 77: 750-753.

BHARGAVA, S., FRISNER, H., BISGARD-FRANTZEN, H. & TAMS, J.W., 2005. Production of ethanol from enzymatically hydrolyzed starch. *Novozymes North America IU, Novozymes A/S*, patent 2005113785.

BHATTACHARYYA, M.K., SMITH, A.M., ELLIS, T.H.N., HEDLEY, C. & MARTIN, C., 1990. The Wrinkled-Seed Character of Pea Described by Mendel Is Caused by a Transposon-Like Insertion in a Gene Encoding Starch-Branching Enzyme. *Cell* 60: 115-122.

BHATTY, R.S. & ROSSNAGEL, B., 1997. Zero amylose lines of hull-less barley. *Cereal Chem*. 74(2): 190-191.

BHAVE, M.R., LAWRENCE, S., BARTON, C. & HANNAH, L., 1990. Identification and molecular characterization of Shrunken-2 cDNA clones of maize. *Plant Cell*. 2: 581-588.

BILIADERIS, C.G., 1991. The structure and interaction of starch with food constituents. *Canadian Journal of Physiology and Pharmacology* 69: 60-78.

BIRD, A.R., BROWN, I.L. & TOPPING, D.L., 2000. Starches, resistant starches, the gut microflora and human health. *Curr. Issues Intest. Microbiol*. 1: 25-37.

BJORCK, I., ELIASSON, A.-C., DREWS, A., GUDMUNDSSON, M. & KARLSSON, R., 1990. Some nutritional properties of starch and dietary fiber in barley genotypes containing different levels of amylose. *Cereal Chem*. 67(4): 327-333.

BLAKE, N.K., SHERMAN, J.D., DVORAK, J. & TALBERT, L.E., 2004. Genome-specific primer sets for starch biosynthesis genes in wheat. *Theor. Appl. Genet*. 109: 1295-1302.

BLAKESLEE, A.F. & AVERY, A.G., 1937. Methods of inducing doubling of chromosomes in plants. *Hered*. 28: 392-411.

BLAUTH, S.L., KIM, K.N., KLUCINEC, J., SHANNON, J.C., THOMPSON, D. & GUILTINAN, M., 2002. Identification of mutator insertional mutants of starch-branching enzyme 1 (sbe1) in *Zea mays* L. *Plant Molecular Biology* 48: 287-297.

BLENNOW, A., BAY-SMIDT, A.M., OLSEN, C.E. & MØLLER, B.L., 2000. The distribution of covalently bound phosphate in the starch granule in relation to starch crystallinity. *Int. J. Biol. Macromol*. 27: 211-218.

BLENNOW, A., NIELSEN, T.H., BAUNSGAARD, L., MIKKELSEN, R. & ENGELSEN, S.B., 2002. Starch phosphorylation: a new front line in starch research. *Trends Plant Sci*. 7: 445-450.

BLOOM, T.M., SYLVESTER-BRADLEY, R., VAIDYANATHAN, L.V. & MURRAY, A.W.A., 1988. Apparent recovery of applied N by winter wheat. In: Jenkinson, D.S. & Smith, K.A. (Eds.), *Efficiency of Nitrogen Use*. Elsevier, London, pp. 27-37.

BLUMENTHAL, C., BEKES, F., GRAS, P.W., BARLOW, E.W.R. & WRIGLEY, C.W., 1995. Identification of wheat genotypes tolerant to the effects of heat stress on grain quality. *Cereal Chem*. 72: 539-544.

BOCCALANDRO, H., PLOSCHUK, E., YANOVSKY, M., SANCHEZ, R., GATZ, C. & CASAL, J., 2003. Increased phytochrome B alleviates density effects on tuber yield of field potato crops. *Plant Physiol*. 133: 1539-1546.

BOLTZ, K.W. & THOMPSON, D.B., 1999. Initial heating temperature and native lipid affects ordering of amylose during cooling of high amylose starches. *Cereal Chem*. 76: 294-212.

BONNER, J.F. & VERNER, J.E., 1976. Plant Biochemistry. 3rd edition, New York, Academic Press, 630 pp.

BOTHAST, R.J. & SCHLICHER, M.A., 2005. Biotechnological processes for conversion of corn into ethanol. *Appl. Microbiol. Biotechnol*. 67: 19-25.

BOWSHER, C.G., TETLOW, I.J., LACEY, A.E., HANKE, G.T. & EMES, M.J., 1996. Integration of metabolism in non-photosynthetic plastids of higher plants. *Compte rendu hebdomadaire des seances de l'Academie des Sciences, Paris* 319: 853-860.

BOYER, C.D., 1976. Changes in starch granule size and amylose percentage during kernel development in several *Zea mays* L. genotypes. *Cereal Chem*. 53(3): 327-337.

BOYER, C.D. & PREISS, J., 1978a. Multiple forms of (1→4)-α-D-glucan,(1→4)-α-D-glucan-6-glycosyl transferase from developing *Zea mays* L. kernels. *Carbohydr. Res*. 61: 321-334.

BOYER, C.D. & PREISS, J., 1978b. Multiple forms of starch branching enzyme in maize: evidence for independent genetic control. *Biochem. Biophys. Res. Commun*. 80: 169-175.

BOYER, C.D. & PREISS, J., 1981. Evidence for independent genetic control of the multiple forms of maize endosperm branching enzymes and starch synthases. *Plant Physiol*. 67: 1141-1145.

BRENNAN, C.S. & CLEARY, L.J., 2005. The potential use of cereal (1-3, 1-4)-b-d-glucans as functional food ingredients. *J. Cereal Sci*. 42: 1-13.

BRIARTY, L.G., HUGHES, C.E. & EVERS, A.D., 1979. The developing endosperm of wheat: a stereological analysis. *Ann. Bot*. 44: 641-658.

BROCKETT, E., THOMPSON, D., DAVIS, T. & BOYER, C., 1988. Gelatinization characteristics of starch from *du*, *wx*, *ae* and *aewx* endosperm of sweet corn inbred Ia 5125. *Starch/Stärke* 40: 175-177.

BROSNAN, J.M., MAKARI, S., PATERSON, L. & COCHRANE, P., 1999. What makes a good grain distillery wheat? In: Campbell, I. (ed.), *Proceedings of the fifth Aviemore conference on malting, brewing and distilling*. Institute of Brewing, London, pp. 225-228.

BROWN, I.L., MCNAUGHT, K.J., ANDREWS, D. & MORITA, T., 2001. Resistant starch: Plant breeding, applications, development and commercial use. In: McCleary, B.V. & Prosky, L. (Eds.). *Advanced dietary fibre technology*. Oxford: Blackwell Science, pp. 401-412.

BRUMOVSKY, J.O. & THOMPSON, D.B., 2001. Production of boiling-stable granular resistant starch by partial acid hydrolysis and hydrothermal treatments of high-amylose maize starch. *Cereal Chem*. 78(6): 680-689.

BRUNNER, B.R. & FREED, R.D., 1994. Oat grain b-glucan content are affected by nitrogen level, location and year. *Crop Sci*. 34: 473-476.

BRUNT, K., 2000. Collaborative Study Concerning the Enzymatic Determination of Starch in Food, Feed, and Raw Materials of the Starch Industry. *Starch/Stärke* 52(2-3): 73-75.

BRUNT, K., 2006. Development and Testing of an Automated Semi-industrial System for in-line NIR Characterization of Potato Composition and Quality. *The 57th Starch Convention*. Retrieved November 16, 2007, from http://www.agfdt.de/loads/st06/brunt.pdf

BRYAN, T., 2002. The search for a perfect ethanol corn. *Ethanol Producer Magazine*, September 2002. Retrieved November 4, 2007, from http://www.ethanolproducer.com/article.jsp?article_id=1619&q=&page=all

BUCKERIDGE, M.S., RAYON, C., URBANOWICZ, B., TINE, M.A.S. & CARPITA, N.C., 2004. Mixed linkage (1-3, 1-4)-b-D-glucans of grasses. *Cereal Chem*. 81: 115-127.

BULEON, A., COLONNA, P., PLANCHOT, V. & BALL, S., 1998. Starch granules: structure and biosynthesis. *Intl. J. Biol. Macromol.* 23: 85-112.

BURGOS-HERNANDEZ, A., ROSAS-BURGOS, C., RAMIREZ-WONG, B., CARBONELL-BARRACHINA, A.A. & CINCO-MOROYOQUI, F.J., 1999. Identification of a-amylase inhibitors in triticale grain. *J. Sci. Food Agric*. 79: 1671-1675.

BURRELL, M.M., 2003. Starch: the need for improved quality or quantity – an overview. *J. Exp. Bot.* 54(382): 451-456.

BURTON, R.A., BEWLEY, J.D., SMITH, A.M., BHATTACHARRYA, M.K., TATGE, H., RING, S., BULL, V., HAMILTON, W.D.O. & MARTIN, C., 1995. Starch branching enzymes belonging to distinct enzyme families are differentially expressed during pea embryo development. *Plant J*. 7: 3-15.

BURTON, R.A., JENNER, H., CARRANGIS, L., FAHY, B., FINCHER, G.B., HYLTON, C., LAURIE, D.A., PARKER, M., WAITE, D., VAN WEGEN, S., VERHOEVEN, T. & DENYER, K., 2002b. Starch granule initiation and growth are altered in barley mutants that lack isoamylase activity. *Plant J.* 31(7): 97-112.

BURTON, R.A., JOHNSON, P.E., BECKLES, D.M., FINCHER, G.B., JENNER, H., NALDRETT, M.J. & DENYER, K., 2002a. Characterization of the genes encoding the cytosolic and plastidial forms of ADP-glucose pyrophosphorylase in wheat endosperm. *Plant Physiol*. 130: 1464-1475.

BUTZEN, S., HAEFELE, D. & HILLARD, P., 2003. Corn processing II: dry grind ethanol production, 13. Pioneer Hybrid International, Johnston.

CAIRNS, J.JR., 2006. Sustainability and the Global Commons. *Asian J. Exp. Sci.* 20(2): 217-224.

CAIRNS, P., BOGRACHEVA, T.YA., RING, S.G., HEDLEY, C.L. & MORRIS, V.J., 1996. Determination of the polymorphic composition of smooth pea starch. *Carbohydr. Polym.* 32: 275-282.

CAMP, W.V., 2005. Yield enhancement genes: seeds for growth. *Current Opinion in Biotechnology* 16: 147-153.

CAMPBELL, M.R., LI, J., BERKE, T.G. & GLOVER, D.V., 1996. Variation of starch granule size in tropical maize germplasm. *Cereal Chem*. 73(5): 536-538.

CAMPBELL, M.R., MANNIS, S.R., PORT, H.A., ZIMMERMAN, A.M. & GLOVER, D.V., 1999. Prediction of Starch Amylose Content Versus Total Grain Amylose Content in Corn by Near-Infrared Transmittance Spectroscopy. *Cereal Chem*. 76(4): 552-557.

CAMPBELL, M.R., SYKES, J. & GLOVER, D.V., 2000. Classification of single and double-mutant corn endosperm genotypes by near-infrared transmittance spectroscopy. *Cereal Chem*. 77: 774-778.

CAO, H., JAMES, M.G. & MYERS, A.M., 2000. Purification and characterization of soluble starch synthases from maize endosperm. *Arch. Biochem. Biophys*. 373: 135-146.

CAO, H. & SHANNON, J.C., 1997. *Bt 1*, a possible adenylate translocator, is developmentally expressed in maize endosperm but not detected in starchy tissue from several other species. *Physiol. Plant.* 100: 400-406.

CAPOUCHOVA, I. & MARESOVA, J.P.D., 2003. Evaluation of size distribution of stach granules in selected wheat varieties by the low angle laser light scattering method. *Plant, Soil and Environment* 49: 12-17.

CARRARI, F., URBANCZYK-WOCHNIAK, E., WILLMITZER, L. & FERNIE, A., 2003. Engineering central metabolism in crop species: learning the system. *Metab. Eng*. 5: 191-200.

CASEY, R., DOMONEY, C. & SMITH, A.M., 1993. Biochemistry and molecular biology of seed products. In: R. Casey, D.R. Davies (Eds.), *Peas: Genetics, Molecular Biology and Biotechnology*. Cambrige: CAB Int., pp. 121-163.

CASPAR, T., 1994. Genetic dissection of the biosynthesis, degradation, and biological functions of starch. In: Meyerowitz E.M., Somerville C.R. (Eds.), *Arabidopsis*. Cold Spring Harbor Laboratory Press, New York, pp. 913-936.

CHAMP, M. & NOAH, L., 1996. Starch bioavailability and glycemic response in humans. *Grain Legumes* 11: 16.

CHAPLIN, M., 2007. Water structure and science: Starch. Retrieved November 28, 2008, from http://www.lsbu.ac.uk/water/hysta.html

CHAWNUA, A., 2000. Development of rapid analytical methods for determining corn quality. Ph.D. abstract, AAT 9991976. The University of Nebraska, Lincoln. 173 pp.

CHEN, M.-H. & BERGMAN, C.J., 2007. Method for determining the amylose content, molecular weights, and weight- and molar-based distributions of degree of polymerization of amylose and fine-structure of amylopectin. *Carbohydr. Polym.* 69: 562-578.

CHEN, M.-H., BERGMAN, C., PINSON, S. & FJELLSTROM, R., 2008. Waxy gene haplotypes: Associations with apparent amylose content and the effect by the environment in an international rice germplasm collection. *J. Cereal Sci.* 47(3): 536-545.

CHEN, C.L. & SUNG, J.M., 1994. Carbohydrate metabolism enzymes in CO_2-enriched developing rice grains varying in grain size. *Physiol. Plant*. 90: 79-85.

CHIOTELLI, E. & LE MESTE, M., 2002. Effect of small and large wheat starch granules on thermomechanical behaviour of starch. *Cereal Chem*. 79: 286-293.

CHOI, S.-B., ZHANG, Y., ITO, H., STEPHENS, K., WINDER, T., EDWARDS, G.E. & OKITA, T.W., 1998. In: Waterlow, J.C. (Ed.), *Feeding a World Population of More than Eight Billion People: A Challenge to Science*. Oxford Univ. Press, Oxford, pp. 137-149.

CHUNG, H.J., SEHNKE, P.C. & FERL, R.J., 1999. The 14-3-3 proteins: cellular regulators of plant metabolism. *Trends Plant Sci.* 4: 367-371.

CIMMYT, 2006. Rust Scoring Guide. Mexico DF, Mexico, CIMMYT. 11 p. Retrieved February 4, 2006, from http://wheatdoctor.cimmyt.org/dmdocuments/rust_scoring_guide.pdf

CLARKE, B.R., DENYER, K., JENNER, C.F. & SMITH, A.M., 1999. The relationship between the rate of starch synthesis, the adenosine 5`-diphosphoglucose concentration and the amylose content of starch in developing pea embryos. *Planta* 209: 324-329.

COGDILL, R.P., HURBURGH, C.R. & RIPPKE, G.R., 2004. Single-kernel maize analysis by near-infrared hyper spectral imaging. *Transactions of the ASAE* 47: 311-320.

COLES, G.D., HARTUNIAN-SOWA, S.M., JAMIESON, P.D. & HAY, A.J., 1997. Environmentally-induced variation in starch and non-starch polysaccharide content in wheat. *Journal of Cereal Science* 26: 47-54.

COLONNA, P., LOUDRIN, D., DELLA VALLE, G. & BULEON, A., 1995. Importance of amylose in non-food uses of pea starches for thermoplastic materials. *2nd Europ. Conf. Grain Legumes*. Copenhagen, pp. 354-355.

COMAI, L. & HENIKOFF, S., 2006. TILLING: practical single-nucleotide mutation discovery. *Plant J.* 45(4): 684-694.

CORNELL, H.J., HOVELING, A.W., CHRYSS, A. & ROGERS, M., 1994. Particle size distribution in wheat starch and its importance in processing. *Starch/Stärke* 46: 203-207.

CORPET, F., GOURY, J. & KAHN, D., 1999. The Pro Dom database of protein domain families. *Nucl. Acids Res*. 26: 323-326.

CRAIG, J., LLOYD, J.R., TOMLINSON, K., BARBER, L., EDWARDS, A., WANG, T.L., MARTIN, C., HEDLEY, C.L. & SMITH, A.M., 1998. Mutations in the gene encoding starch synthase II profoundly alter amylopectin structure in pea embryos. *The Plant Cell*. 10: 413-426.

CROSSA, J. & CORNELIUS, P.L., 1997. Sites regression and shifted multiplicative model clustering of cultivar trial sites under heterogeneity of error variances. *Crop Sci.* 37: 405-415.

CROSSA, J., CORNELIUS, P.L. & YAN, W., 2002. Biplots of linear-bilinear models for studying crossover genotype x environment interaction. *Crop Sci.* 42: 619-633.

CROWE, T.C., SELIGMAN, S.A. & COPELAND, L., 2000. Inhibition of enzymic digestion of amylose by free fatty acids in vitro contributes to resistant starch formation. *J. Nutr*. 130: 2006-2008.

CUI, R. & OATES, C.G., 1999. The effect of amylose-lipid complex formation on enzyme susceptibility of sago starch. *Food Chem*. 65: 417-425.

CURRAN, P.J., DUNGAN, J.L., MACLER, B.A., PLUMMER, S.E. & PETERSON, D.L., 1992. Reflectance spectroscopy of fresh whole leaves for the estimation of chemical concentration. *Remote Sensing of Environment* 39(2): 153-166.

CYRAN, M., RAKOWSKA, M. & MIAZGA, D., 1996. Chromosomal location of factors affecting content and composition of non-starch polysaccharides in wheat-rye addition lines. *Euphytica* 89(1): 153-157.

DAI, Z.-M., YIN, Y.-P., ZHANG, M., LI, W.-Y., YAN, S.-H., CAI, R.-G. & WANG, Z.-L., 2008. Distribution of Starch Granule Size in Grains of Wheat Grown Under Irrigated and Rainfed Conditions. *Acta Agron. Sin.* 34(5): 795-802.

DANG, P.L. & BOYER, C.D., 1988. Maize leaf and kernel starch synthases and starch branching enzymes. *Phytochemistry* 27: 1255-1259.

DAVIS, J.P., SUPATCHAREE, N., KHANDELWAL, R.L. & CHIBBAR, R.N., 2003. Synthesis of Novel Starches in Planta: Opportunities and Challenges. *Starch/Stärke* 55(3-4): 107-120.

DAVIS-KNIGHT, H.R. & WEIGHTMAN, R.M., 2008. The potential of triticale as a low input cereal for bioethanol production. *HGCA Project No. 3348, Project Report No. 434*, July 2008, 41 p.

DEAT RSA, 2008. The Emergence of a 'Green Fuel' in South Africa - Biofuels: A Consideration of Key Emerging Issues that may Impact the State of the Environment. *Emerging Issues Paper: Biofuels*. Paper prepared by Dr Haywood, L. (CSIR, Natural Resources and the Environment) for the Department of Environmental Affairs and Tourism (DEAT), Republic of South Africa. March 2008, 11 pp.

DEBON, S.J.J., TESTER, R.F., MILLAM, S. & DAVIES, H.V., 1998. Effects of temperature on the synthesis, composition and physical properties of potato microtuber starch. *J. Sci. Food Agr.* 76(4): 599-607.

DEFRA, 2004. Genetic Reduction of Energy use and Emissions of Nitrogen in cereal production, GREEN grain (LK0959). *DEFRA Science and Research*. Retrieved July 3, 2006, from http://www2.defra.gov.uk/research/Project_Data/More.asp?I=LK0959&M=CFO&V=

DELWICHE, S.R., 1995. Single wheat kernel analysis by near-infrared transmittance: protein content. *Cereal Chem.* 72: 11-16.

DELWICHE, S.R., 1998. Protein content of single kernels of wheat by nearinfrared spectroscopy. *J. Cereal Sci*. 27: 241-254.

DELWICHE, S.R., BEAN, M.M., MILLER, R.E., WEBB, B.D. & WILLIAMS, P.C., 1995. Apparent amylose of milled rice by near-infrared reflectance spectrophotometry. *Cereal Chem.* 72: 182-187.

DELWICHE, S.R. & GRAYBOSCH, R.A., 2002. Identification of Waxy Wheat by Near-infrared Reflectance Spectroscopy. *J. Cereal Sci*. 35: 29-38.

DELWICHE, S.R., GRAYBOSCH, R.A., HANSEN, L.E., SOUZA, E. & DOWELL, F.E., 2006. Single Kernel Near-Infrared Analysis of Tetraploid (Durum) Wheat for classification of the waxy condition. *Cereal Chem.* 83(3): 287-292.

DELWICHE, S.R. & HRUSCHKA, W.R., 2000. Protein content of bulk wheat from near-infrared reflectance of individual kernels. *Cereal Chem.* 77: 86-88.

DELWICHE, S.R. & MASSIE, D.R., 1996. Classification of Wheat by Visible and Near-Infrared Reflectance from Single Kernels. *Cereal Chem*. 73(3): 399-405.

DELWICHE, S.R., MCKENZIE, K.S. & WEBB, B.D., 1996. Quality Characteristics in Rice by Near-Infrared Reflectance Analysis of Whole-Grain Milled Samples. *Cereal Chem*. 73(2): 257-263.

DEMEKE, T., HUCL, P., ABDEL-AAL, E.S.M., BÅGA, M. & CHIBBAR, R.N., 1999. Biochemical characterization of the wheat waxy A protein and its effect on starch properties. *Cereal Chem*. 76: 69-77.

DEMIATE, I.M., KONKEL, F.E. & PEDROSO, R.A., 2001. Enzymatic determination of starch in Doce de Leite using dialysis. *Cienc. Tecnol. Aliment. Campinas* 21(3): 339-342.

DENYER, K., BARBER, L.M., BURTON, R., HEDLEY, C.L., HYLTON, C.M., JOHNSON, S., JONES, D.A., MARSHALL, J., SMITH, A.M., TATGE, H., TOMLINSON, K. & WANG, T.L., 1995. The isolation and characterization of novel, low-amylose mutants of *Pisum sativum* L. *Plant Cell Environ*. 18: 1019-1026.

DENYER, K., BARBER, L.M., EDWARDS, E.A., SMITH, A.M. & WANG, T.L., 1997a. Two isoforms of the GBSS I class of granule-bound starch synthase are differentially expressed in the pea plant. *Plant Cell Environ*. 20: 1566-1572.

DENYER, K., CLARKE, B., HYLTON, C., TATGE, H. & SMITH, A.M., 1996. The elongation of amylase and amylopectin chain in isolated starch granules. *Plant J.* 10: 1135-1143.

DENYER, K., DUNLAP, F., THORBJORNSEN, T., KEELING, P. & SMITH, A.M., 1996. The major form of ADP-glucose pyrophosphorylase in maize endosperm is extra-plastidial. *Plant Physiol*. 112: 779-785.

DENYER, K., EDWARDS, A., MARTIN, C. & SMITH, A.M., 1997b. The mechanism of amylose synthesis. *Starch: structure and functionality*. The Royal Society of Chemistry, Cambridge, UK, pp. 222-229.

DENYER, K., HYLTON, C.M., JENNER, C.F. & SMITH, A.M., 1995. Identification of multiple isoforms of soluble and granule-bound starch synthase in developing wheat endosperm. *Planta* 196: 256-265.

DENYER, K., JOHNSON, P., ZEEMAN, S. & SMITH, A.M., 2001. The control of amylose synthesis. *J. Plant Physiol*. 158: 479-487.

DENYER, K., SIDEBOTTOM, C., HYLTON, C.M. & SMITH, A.M., 1993. Soluble isoforms of starch synthase and starch-branching enzyme also occur within starch granules in developing pea embryos. *Plant J*. 4(2): 191-198.

DIAN, W.M., JIANG, H.W. & WU, P., 2005. Evolution and expression analysis of starch synthase III and IV in rice. *J. Exp. Bot.* 56: 623-632.

DINGES, J.R., COLLEONI, C., JAMES, M.G. & MYERS, A.M., 2003. Mutational analysis of the pullulanase-type debranching enzyme of maize indicates multiple functions in starch metabolism. *Plant Cell*. 15: 666-680.

DINGES, J.R., COLLEONI, C., MYERS, A.M. & JAMES, M.G., 2001. Molecular structure of three mutations at the maize sugary1 locus and their allele-specific phenotypic effects. *Plant Physiol*. 125: 1406-1418.

DME RSA, 2006. Draft Biofuels Industrial Strategy of the Republic of South Africa. *Department of Minerals and Energy, Republic of South Africa*. November 2006, 16 pp.

DME RSA, 2007. Biofuels Industrial Strategy of the Republic of South Africa. *Department of Minerals and Energy, Republic of South Africa*. December 2007, 29 pp.

DOAN, D.N., RUDI, H. & OLSEN, O.A., 1999. The allosterically unregulated isoform of ADP-glucose pyrophosphorylase from barley endosperm is the most likely source of ADP-glucose incorporated into endosperm starch. *Plant Physiol*. 121: 965-975.

DOEHLERT, D. & KNUTSON, C.A. 1991. Two classes of starch debranching enzymes from developing maize kernels. *J. Plant Physiol*. 138: 566-572.

DOLL, H., 1983. Barley seed proteins and possibilities for their improvement. In: Gottchalk, W., Muller, H.P. (Eds.), *Seed Proteins*. Martinus Nijhoff/W. Junk, The Hague, pp. 205-223.

DOMBRINK-KURTZMAN, M.A. & KNUTSON, C.A., 1997. A study of maize endosperm hardness in relation to amylose content and susceptibility to damage. *Cereal Chem.* 74: 776-780.

DONALD, A.M., 2001. Plasticization and self-assembly in the starch granule. Review. *Cereal Chem*. 78: 307-314.

DOWELL, F.E., MAGHIRANG, E.B., GRAYBOSCH, R.A., BAENZIGER, P.S., BALTENSPERGER, D.D. & HANSEN, L.E., 2006b. An Automated Near-Infrared System for Selecting Individual Kernels Based on Specific Quality Characteristics. *Cereal Chem.* 83(5): 537-544.

DOWELL, F.E., MAGHIRANG, E.B., XIE, F., LOOKHART, G.L., PIERCE, R.O., SEABOURN, B.W., BEAN, S.R., WILSON, J.D. & CHUNG, O.K., 2006a. Predicting Wheat Quality Characteristics and Functionality Using Near-Infrared Spectroscopy. *Cereal Chem.* 83(5): 529-536.

DOWELL, F.E., PEARSON, T.C., MAGHIRANG, E.B., XIE, F. & WICKLOW, D.T., 2002. Reflectance and transmittance spectroscopy applied to detecting fumonisin in single corn kernels infected with Fusarium verticillioides. *Cereal Chem.* 79: 222-226.

DRY, I., SMITH, A., EDWARDS, A., BHATTACHARYYA, M., DUNN, P. & MARTIN, C., 1992. Characterization of cDNA encoding two isoforms of granule-bound starch synthase with show differential expression in developing storage organs of pea and potato. *Plant J.* 2: 193-202.

DU PISANI, F., 2009. Evaluation of the structural and functional composition of South African triticale cultivars (×*Triticosecale* Wittmack). Thesis presented in partial fulfilment of the requirements for the degree of Master of Science in Food Science. Department of Food Science, Stellenbosch University, March 2009. 141 pp.

DUKE, E.R. & DOEHLERT, D.C., 1996. Effects of heat stress on enzyme activities and transcript levels in developing maize kernels grown in culture. *Environ. Exp. Bot.* 36(2): 199-208.

DUWENIG, E., STEUP, M. & KOSSMANN, J., 1997. Induction of genes encoding plastidic phosphorylase from spinach and potato by exogenously supplied carbohydrates in excised leaf discs. *Planta* 203: 111-120.

EDWARDS, A., VINCKEN, J.-P., SUURS, L.C.J.M., R. VISSER, G.F., ZEEMAN, S., SMITH, A. & MARTIN, C., 2002. Discrete forms of amylose are synthesized by isoforms of GBSSI in pea. *Plant Cell*. 14(8): 1767-1785.

ELIASSON, A.-C. & LARSSON, K., 1993. Cereals in Breadmaking. New York, Marcel Dekker, Inc., 376 pp.

ELLIS, R.P., COCHRANE, M.P., DALE, M.F.B., DUFFUS, C.M., LYNN, A., MORRISON, I.M., PRENTICE, R.D.M., SWANSTON, J.S. & TILLER, S.A., 1998. Starch production and industrial use. *J. Sci. Food Agric*. 77: 289-311.

ENGLYST, H.N., QUIGLEY, M.E., HUDSON, G.J. & CUMMINGS, J.H., 1992. Determination of dietary fibre as non starch polysaccharides by gas-liquid chromatography. *Analyst* 117: 1707-1714.

EREKUL, O. & KOHN, W., 2006. Effect of Weather and Soil Conditions on Yield Components and Bread-Making Quality of Winter Wheat (*Triticum aestivum* L.) and Winter Triticale (*Triticosecale* Wittm.) Varieties in North-East Germany. *J. Agron. Crop Sci.* 192: 452-464.

ESBENSEN, K.H., 2002. Multivariate data analysis – in practice. 5th edition, CAMO Process AS, Oslo, Norway, 598 pp.

ETTER, L., 2007. Power plant in China: a plan to turn rice into carbon credits. *Wall Street Journal*, 9 October 2007. Retrieved November 10, 2007, from http://online.wsj.com/public/article/SB119187524509952568.html

EVANS, A. & THOMPSON, D.B., 2004. Resistance to [alpha]-Amylase Digestion in Four Native High-Amylose Maize Starches. *Cereal Chem.* 81(1): 31-37.

EVERS, A.D. & LINDLEY, J., 1977. The particle-size distribution in wheat endosperm starch. *J. Sci. Food Agric*. 28: 98-101.

EZEOGU, L.I., DUODU, K.G. & TAYLOR, J.R.N., 2005. Effects of endosperm texture and cooking conditions on the in vitro starch digestibility of sorghum and maize flours. *J. Cereal Sci*. 42: 33-44.

FALK, A., SCHULMAN, A.H., RASMUSSEN, S. & JANSSON, C., 2001. Barley mutagenesis. In: Karl Esser (Ed.), *Progress in Botany*. Springer-Verlag, Berlin, pp. 32-50.

FANNON, J.E., HUBER, R.J. & BEMILLER, J.N., 1992. Surface pores of starch granules. *Cereal Chem.* 69: 284-288.

FANNON, J.E., HUBER, R.J. & BEMILLER, J.N., 1993. Inferior channels of starch granule. *Cereal Chem*. 70: 611-613.

FAO, 1998. Carbohydrates in human nutrition. *FAO Food and Nutrition Paper - 66. Report of a Joint FAO/WHO Expert Consultation.* Rome, 14-18 April 1997. FAO, Viale delle Terme di Caracalla, 00100 Rome, Italy. Retrieved February 28, 2007, from http://www.fao.org/docrep/W8079E/w8079e00.HTM

FARADZHEVA, E.D., 2000. Vliyanie ras drozhzhej na obrazovanie vysshikh spirtov i diatsetila v proizvodstve piva (in Russian). *Vestnik VGTA* 5: 38-43.

FEIZ, L., MARTIN, J.M. & GIROUX, M.J., 2008. Relationship between Wheat (*Triticum aestivum* L.) Grain Hardness and Wet-Milling quality. *Cereal Chem.* 85(1): 44-50.

FELL, D.A., 1999. Traditional concepts of metabolic control mislead more than enlighten. *The Biochemist*, February 1999, 13-16.

FERGASON, V.L., 1994. High amylose and waxy corns. In: A. R. Hallauer (ed.), *Specialty Corns*. CRC Press Ltd, Boca Raton, FL, USA, pp. 55-57.

FERGASON, V.L. & ZUBER, M.S., 1962. Influence of environment on amylose content of maize endosperm. *Crop Sci*. 2(2): 209-211.

FERGASON, V.L. & ZUBER, M.S., 1965. Response of high amylose maize hybrids to environmental influences. *Crop Sci*. 5(2): 169-170.

FERTIG, C.C., PODCZECK, F., JEE, R.D. & SMITH, M.R., 2004. Feasibility study for the rapid determination of the amylose content in starch by near-infrared spectroscopy. *European Journal of Pharmaceutical Sciences* 21: 155-159.

FISHER, D.K., BOYER, C.D. & HANNAH, L.C., 1993. Starch branching enzyme II from maize endosperm. *Plant Physiol*. 102: 1045-1046.

FISHER, L.R., CARRINGTON, S.P. & ODELL, J.A., 1997. Deformation mechanics of individual swollen starch granules. In: Frazier, P.J., Donald, A.M., Richmond, P. (Eds.), *Starch: Structure and Functionality*. The Royal Society of Chemistry, London, UK, pp. 105-114.

FISHER, D.K., GAO, M., KIM, K.-N., BOYER, C.D. & GUITINAN, M.J., 1996. Allelic analysis of the maize (*Zea mays* L.) amylose-extender locus suggests that independent genes encode starch branching enzymes IIa and IIb. *Plant Physiol*. 110: 611-619.

FISHER, D.K., KIM, K.-N., GAO, M., BOYER, C.D. & GUITINAN, M.J., 1995. A cDNA encoding starch branching enzyme I from maize endosperm. *Plant Physiol*. 108: 1313-1314.

FISHER, D.K. & THOMPSON, D.B., 1997. Retrogradation of maize starch after thermal treatment within and above the gelatinization temperature grade. *Cereal Chem*. 74: 344-351.

FLEISCHER, S. & SENN, T., 2005. Regional plants – their potential for sustainable ethanol production, their ecobalance and the effects of cereal properties on ethanol yields. *Zuckerindustrie* 130: 773-777.

FLINTHAM, J., EVERS, A.D. & KRATOCHVIL, J., 1993. Genetical and physiological studies on sprouting in Wheat. *HGCA, Project Report 77*, London.

FLIPSE, E., KEETELS, C.J.A.M., JACOBSON, E. & VISSER, R.G.F., 1996. The dosage effect of the wild type GBSS allele is linear for GBSS activity but not for amylase content: absence of amylose has a distinct influence on the physico-chemical properties of starch. *Theor. Appl. Genet*. 92: 121-127.

FORTUNA, T., GAMBUS, H., NOWOTNA, A. & PALASINSKI, M., 1985. Studies on the suitability of triticale for starch production. *Acta Aliment. Pol*. 11(35)1: 53-62.

FORWARD, P., 1994. Beyond ethanol: Industrial uses of agricultural materials. *A background and opportunities paper*. Food Bureau, Market and Industry Services Branch, Agriculture and Agri-Food Canada. 55 pp.

FOSS, 2008. Infratec™ 1241 Systems offer simultaneous determinations of protein, moisture, fat, fibre, starch, amylose, colour, oil and volume weight in grains – without sample preparation – in less than one minute. *FOSS*. Retrieved November 4, 2007, from http://www.foss.dk/Solutions/ProductsDirect/Infratec1241GrainAnalyzer.aspx

FOULKES, M.J., SYLVESTER-BRADLEY, R. & SCOTT, R.K., 1998. Evidence for differences between winter wheat cultivars in aquisition of soil mineral nitrogen and uptake and utilization of applied fertilizer nitrogen. *J. Agric. Sci.* 130: 29-44.

FREEMAN, J.E., BOCAN, B.J. & ZOBEL, H.F., 1972. Starch: variation associated with location in corn and sorghum plants. *Crop Sci*. 12(1): 122-124.

FRENCH, D., 1984. Organization of starch granule. In: R.L. Whistler, J.N. BeMiller, E.F. Parshall (Eds.), *Starch: Chemistry and technology*. Academic Press, Orlando, FL, pp. 183-247.

FUJITA, N. & TAIRA, T., 1998. A 56-kDa protein is a novel granulo-bound starch synthase existing in the pericarps, aleurone layers, and embryos of immature seed in diploid wheat (*Triticum monococcum* L.). *Planta* 207: 125-132.

FUJITA, N., WADANO, A., KOZAKI, S., TAKAOKA, K., OKABE, S. & TAIRA, T., 1996. Comparison of the primary structure of waxy protein (granule-bound starch synthase) between polyploidy wheats and related diploid species. *Biochemical Genetics* 34: 403-413.

FUJITA, S., YAMAMOTO, H., SUGIMOTO, Y., MORITA, N. & YAMAMORI, M., 1998. Thermal and crystalline properties of waxy wheat (*Triticum aestivum* L.) starch. *J. Cereal Sci*. 27: 1-5.

FUJITA, N., YOSHIDA, M., ASAKURA, N., OHDAN, T., MIYAO, A., HIROCHIKA, H. & NAKAMURA, Y., 2006. Function and characterization of starch synthase I using mutants in rice. *Plant Physiol.* 140: 1070-1084.

FUWA, H., TAKAYA, T. & SUGIMOTO, Y., 1980. Degradation of various starch granules by amylases. In: Marshall (ed.), *Mechanisms of Saccharide Polymerization and Depolymerization*, 3.3. Academic Press, New York, NY, pp. 73-102.

GAINES, C.S., RAEKER, M.O. & TILLEY, M., 2000. Associations of starch gel hardness, granule size, waxy allelic expression, thermal pasting, milling quality, and kernel texture of 12 soft wheat cultivars. *Cereal Chem*. 77: 163-168.

GALE, M.D., LAW, C.N., CHOJECKI, A.J. & KEMPTON, R.A., 1983. Genetic control of alpha-amylase production in wheat. *Theor. Appl. Genet.* 64: 309-316.

GALLANT, D.J., BOUCHET, B. & BALDWIN, P.M., 1997. Microscopy of starch: Evidence of a new level of granule organization. *Carbohydr. Polym.* 32: 177-191.

GAO, M., FISHER, D.K., KIM, K.-N., SHANNON, J.C. & GUITINAN, M.J., 1996. Evolutionary conservation and expression patterns of maize starch branching enzyme I and IIb genes suggests isoform specialization. *Plant Mol. Biol*. 30: 1223-1232.

GAO, M., FISHER, D.K., KIM, K.-N., SHANNON, J.C. & GUITINAN, M.J., 1997. Independent genetic control of maize starch-branching enzymes IIa and IIb. *Plant Physiol*. 114: 69-78.

GAO, M., WANAT, J., STINARD, P, JAMES, M. & MYERS, A., 1998. Characterisation of dull 1, a maize gene coding for a novel starch synthase. *Plant Cell*. 10: 399-415.

GARWOOD, D.L., SHANNON, J.C. & CREECH, R.G., 1976. Starches of endosperms possessing different alleles at the amylose extender locus in *Zea mays* L. *Cereal Chem*. 53(4): 355-364.

GEERA, B.P., NELSON, J.E., SOUZA, E. & HUBER, K.C., 2005a. Granule bound starch synthase I (GBSSI) gene effects related to soft wheat flour/starch characteristics and properties. *Cereal Chem*. 83: 544-550.

GEERA, B.P., NELSON, J.E., SOUZA, E. & HUBER, K.C., 2005b. Flour pasting properties of wild-type and partial waxy soft wheats in relation to growing environment-induced fluctuations in starch characteristics. *Cereal Chem*. 83: 558-564.

GEERA, B.P., NELSON, J.E., SOUZA, E. & HUBER, K.C., 2006. Composition and Properties of A- and B-Type Starch Granules of Wild-Type, Partial Waxy, and Waxy Soft Wheat. *Cereal Chem.* 83(5): 551-557.

GEIGER, D.R., KOCH, K.E. & SHIEH, W., 1996. Effect of environmental factors on whole plant assimilate partitioning and associated gene expression. *J. Exp. Bot.* 47: 1229-1238.

GENENCOR, 2005a. Genencor launches industry transforming technology for ethanol production. *Press Release*. Genencor International. Retrieved November 4, 2007, from http://www.genencor.com/wt/gcor/pr_1120661186

GENENCOR, 2005b. Transforming the Ethanol Industry. A General Backgrounder. Genencor International, June 2005. Retrieved November 4, 2007, from http://www.genencor.com/cms/resources/file/ebf95c076d3afc7/STARGEN%20Backgrounder.pdf

GENENCOR, 2006. Genencor launches improved acid fungal protease for use in the ethanol industry. *Press Release*. Genencor International. Retrieved November 4, 2007, from http://www.genencor.com/wt/gcor/pr_1150717658

GENENCOR, 2007. Enzyme Products for Fuel Ethanol Production. Danisco US Inc., April 2007. Retrieved November 4, 2007, from http://www.genencor.com/cms/resources/file/eb03b408b6151cc/GENC-7997%20Eth%20Prod%20List%20Update%20prt%20(april%2007).pdf

GERARD, C., BARRON, C., COLONNA, P. & PLANCHOT, V., 2001. Amylose determination in genetically modified starches. *Carbohydr. Polym.* 44: 19-27.

GERARD, C., COLONNA, P., BULEON, A. & PLANCHOT, V., 2001. Amylolysis of maize mutant starches. *J. Sci. Food Agric.* 81: 1281-1287.

GERNAT, C., RADOSTA, S., ANGER, H. & DAMASCHUN, G., 1993. Crystalline parts of three different conformations detected in native and enzymatically degraded starches. *Starch/Stärke* 45: 309-314.

GIBBON, B.C., WANG, X. & LARKINS, B.A., 2003. Altered starch structure is associated with endosperm modification in quality protein maize. *Proc. New-York Acad. Sci*. 100(26): 15329-15334.

GIBSON, T.S., SOLAH, V. & MCCLEARY, B.V., 1997. A procedure to measure amylose in cereal starches and flours with concanavalin A. *J. Cereal Sci*. 25: 111-119.

GIDLEY, M.J. & BOCIEK, S., 1985. Molecular organization in starches a13 C CP/MAS NMR study. *J. Amer. Chem. Soc*. 107: 7040-7044.

GIROUX, M.J., SHAW, J., BARRY, G., COBB, B.G., GREENE, T., OKITA, T. & HANNAH, L.C., 1996. A single gene mutation that increases maize seed weight. *Proc. Natl. Acad. Sci. USA* 93: 5824-5829.

GLATTHAR, J., HEINISCH, J.J. & SENN, T., 2004. Unmalted triticale cultivars as brewing adjuncts: effects of enzyme activities and composition on beer wort quality. *J. Sci. Food Agric.* 85(4): 647-654.

GLENNA, L.L. & CAHOY, D.R., 2009. Agribusiness concentration, intellectual property, and the prospects for rural economic benefits from the emerging biofuel economy. *Southern Rural Sociology* 24(2): 111–129.

GONCHARENKO, A.A. & TIMOSHCHENKO, A.S., 2006. The relation between the structure of starch complex in winter rye and the viscosity of water extract. (Abstract). Doklady Rossiiskoi Akademii Sel'skokhozyaistvennykh Nauk (2006) No. 6, 6-10 (in Russian). *Russian Agricultural Sciences* 11: 6-10.

GOOD, A.G., JOHNSON, S.J., DE PAUW, M., CARROLL, R.T., SAVIDOV, N., VIDMAR, J., LU, Z., TAYLOR, G. & STROEHER, V., 2007. Engineering nitrogen use efficiency with alanine aminotransferase. *Canadian Journal of Botany* 85: 252-262.

GOOD, A.G., SHRAWAT, A.K. & MUENCH, D.G., 2004. Can less yield more? Is reducing nutrient input into the environment compatible with maintaining crop production? *Trends Plant Sci.* 9(12): 597-605.

GOVERNMENT OF ALBERTA, 2006. Triticale Grain for Other Uses. Government of Alberta, Canada. Retrieved November 4, 2007, from http://www1.agric.gov.ab.ca/$department/deptdocs.nsf/all/fcd10568

GRAHAM, R., LIEW, M., MEADOWS, C., LYON, E. & WITTWER, C.T., 2005. Distinguishing different DNA heterozygotes by high-resolution melting. *Clinical Chem.* 51: 1295-1298.

GRAIN SA, 2006. Grane vir brandstof! Grains for fuel! *SA Graan / Grain Article* 4, Maart 2006. Retrieved November 4, 2007, from http://www.graansa.co.za/documents/SAGraan_Mrt06_P4.pdf

GRANT, L.A., OSTENSON, A.M. & RAYAS-DUARTE, P., 2002. Determination of amylose and amylopectin of wheat starch using high performance size-exclusion chromatography (HPSSEC). *Cereal Chem.* 79: 771-773.

GRAY, J.A. & BEMILLER, J.N., 2004. Development and utilization of reflectance confocal laser scanning microscopy to locate reaction sites in modified starch granules. *Cereal Chem.* 81: 278-286.

GRAY, K.A, ZHAO, L. & EMPTAGE, M., 2006. Bioethanol. *Current Opinion in Chemical Biology* 10: 141-146.

GRAYBOSCH, R.A., 1998. Waxy wheats: Origin, properties, and prospects. *Trends Food Sci. Technol.* 9: 135-142.

GRAYBOSCH, R.A., GUO, G. & SHELTON, D.R., 2000. Aberrant falling numbers of waxy wheats independent of alpha-amylase activity. *Cereal Chem*. 77: 1-3.

GRAYBOSCH, R.A., SOUZA, E., BERZONSKY, W., BAENZIGER, P.S. & CHUNG, O., 2003. Functional properties of waxy wheat flours: Genotypic and environmental effects. *J. Cereal Sci*. 38: 69-76.

GREENE, T. & HANNAH, H., 1998a. Enhanced stability of maize endosperm ADP-glucose pyrophosphorylase is gained through mutants that alter subunit interactions. *Proc. Natl. Acad. Sci. USA* 95: 13342-13347.

GREENE, T.W. & HANNAH, L.C., 1998b. Adenosine diphosphate glucose pyrophosphorylase, a rate-limiting step in starch biosynthesis. *Physiol. Plant*. 103(4): 574-580.

GREENWELL, P. & SCHOFIELD, J.D., 1989. The chemical basis of grain hardness and softness. In: Salovaara, H., (Ed.), *Wheat End-Use Properties*. Proc. ICC Symp. University of Helsinki, Helsinki, Finland, pp. 59-72.

GRESSEL, J., 2008. Transgenics are imperative for biofuel crops. *Plant Sci.* 174: 246-263.

GRIFFIN, G.J.L., 1989. Wheat Starch in the Formulation of Degradable Plastics. In: Y. Pomeranz (Ed.), *Wheat is Unique*. American Assoc. Cereal Chemists, St. Paul, MN, pp. 695-706.

GROOS, C., BERVAS, E. & CHARMET, G., 2004. Genetic analysis of grain protein content, grain hardness and dough rheology in a hardXhard bread wheat progeny. *J. Cereal Sci.* 40: 93-100.

GUAN, H.P. & KEELING, P.L., 1998. Starch biosynthesis: understanding the functions and interactions of multiple isozymes of starch synthase and branching enzyme. *Trends Glicosci. Glycotech.* 10: 307-319.

GUAN, H.P. & PREISS, J., 1993. Differentiation of the properties of the branching isozymes from maize (*Zea mays* L.). *Plant Physiol*. 102: 1269-1273.

GUEDES-PINTO, H., DARVEY, N. & CARNIDE, V.P. (Eds.), 1996. Triticale: Today and Tomorrow. Boston/London, 912 pp.

GUILBOT, A. & MERCIER, C., 1985. Starch. In: G.O. Aspinall (Ed.), *The Polysaccharides.* London: Academic Press, pp. 209-282.

GURAYA, H.S., KADAN, R.S. & CHAMPAGNE, E.T., 1997. Effect of ricestarch lipid complexes on in vitro digestibility complexing index and viscosity. *Cereal Chem.* 74: 561-565.

GUTIERREZ, O.A., CAMPBELL, M.R. & GLOVER, D.V., 2002. Starch particle volume in single- and double mutant maize endosperm genotypes involving the soft starch (h) gene. *Crop Sci*. 42(3): 355-359.

HAARD, N.F., ODUNFA, S.A., LEE, C.-H., QUINTERO-RAMÍREZ, R., LORENCE-QUIÑONES, A. & WACHER-RADARTE, C., 1999. Fermented Cereals. A Global Perspective. *FAO Agricultural Services Bulletin No. 138*. Food and Agriculture Organization of the United Nations, Rome. Retrieved November 28, 2007, from http://www.fao.org/docrep/x2184e/x2184e00.HTM

HACKETT, R. & BURKE, J.I., 2004. Potential for triticale in low cost production systems. TEAGASC, Crops Research Centre, Oak Park, Ireland. Retrieved November 28, 2007, from http://www.teagasc.ie/publications/2004/20040128/paper7.htm

HAMADA, S., NOZAKI, K., ITO, H., YOSHIMOTO, Y., YOSHIDA, H., HIRAGA, S., ONODERA, S., HONMA, M., TAKEDA, Y. & MATSUI, H., 2001. Two starch-branching enzyme isoforms occur in different fractions of developing seeds of kidney bean. *Biochem. J.* 359: 23-34.

HAN, X-Z. & HAMAKER, B.R., 2001. Amylopectin fine structure and rice starch paste breakdown. *J. Cereal Sci*. 34: 279-284.

HANNAH, L.C., GIROUX, M. & BOYER, C.D., 1993. Biotechnological modification for sweet corn and maize improvement. *Sci. Hortic*. 55: 177-197.

HAUSLER, R., HIRXH, H., KREUZALER, F. & PETERHANSEL, C., 2002. Overexpression of C4-cycle enzymes in transgenic C3 plants: a biotechnological approach to improve C3-photosynthesis. *J. Exp. Bot*. 53: 591-607.

HAYAKAWA, K., TANAKA, K., NAKAMURA, T., ENDO, S. & HOSHINO, T., 1997. Quality characteristics of waxy hexaploid wheat (*Triticum aestivum* L.): Properties of starch gelatinization and retrogradation. *Cereal Chem*. 74: 576-580.

HAYAKAWA, K., TANAKA, K., NAKAMURA, T., ENDO, S. & HOSHINO, T., 2004. End use quality of waxy wheat flour in various grain-based foods. *Cereal Chem.* 81: 666-672.

HAYASHI, M., YASUI, T., KIRIBUCHI-OTOBE, C. & SEGUCHI, M. 2004. Presence of a minor amylose fraction in the central portion of waxy wheat starch granules and its contribution to granular stability. *Cereal Chem*. 81: 589-593.

HEATON, E., LONG, S.P., VOIGT, T.B., JONES, M.B. & CLIFTON-BROWN, J., 2004. *Miscanthus* for Renewable Energy Generation: European Union Experience and Projections for Illinois. *Mitigation and Adaptation Strategies for Global Change* 9(4): 433-451.

HEDLEY, C.L., BOGRACHEVA, T.Ya., LLOYD, J.R. & WANG, T.L., 1996. Manipulation of starch composition and quality in pea seeds. In: G.R. Fenwick, C. Hedley, R.L. Richardson, S. Khokhar (Eds.), *Agri-Food Quality: An Interdisciplinary Approach.* London: Royal Soc. Chem., pp. 138-148.

HENCKE, J.M.M., KLEPPER, G. & SCHMITZ, N., 2005. Tax exemption for biofuels in Germany: is bio-ethanol really an option for climate policy. *Energy* 14: 2617-2635.

HEYER, A., RAAP, M., SCHROEER, B., MARTY, B. & WILLMITZER, L., 2004. Cell wall invertase expression at the apical meristem alters floral, architectural, and reproductive traits in Arabidopsis thaliana. *Plant J.* 39: 161-169.

HILLS, M.J., 2004. Control of storage-product synthesis in seeds. *Current Opinion in Plant Biology* 7: 302-308.

HIRANO, H.Y., EIGUCHI, M. & SANO, Y., 1998. A single base change altered the regulation of the Waxy gene at the posttranscriptional level during the domestication of rice. *Molecular Biology and Evolution* 15(8): 978-987.

HIRANO, H. & SANO, Y., 1991. Molecular characterization of the waxy locus of rice (*Oriza sativa*). *Plant Cell. Phisiol*. 32: 989-997.

HIROSE, T. & TERAO, T., 2004. A comprehensive expression analysis of the starch synthase gene family in rice (*Oryza sativa* L.). *Planta* 220: 9-16.

HIUKKA, R., 1998. A multivariate approach to the analysis of pine needle samples using NIR. *Chemometrics and Intelligent Laboratory Systems* 44: 395-401.

HIZUKURI, S., 1969. The effect of environment temperature of plants on the physico-chemical properties of their starches. *J. Jpn. Starch Sci*. 17(1): 73-88.

HIZUKURI, S., 1985. Relationship between the distribution of the chain length of amylopectin and the crystalline structure of starch granules. *Carbohydr. Res.* 141: 295-306.

HIZUKURI, S., 1986. Polymodal distribution of the chain lengths of amylopectins and its significance. *Carbohydr. Res.* 147: 342-347.

HIZUKURI, S., 1996. Starch: analytical aspects. In: Eliasson, A.-C. (Ed.), *Carbohydrates in Food*. Marcel Dekker, New York, pp. 347-429.

HIZUKURI, S., TAKEDA, Y. & YASUDA, M., 1981. Multi-branched nature of amylose and the action of debranching enzymes. *Carbohydr. Res*. 94: 205-213.

HOLM, J., BJORCK, I., DREWS, A. & ASP, N.-G., 1986. A rapid method for the analysis of starch. *Starch/Stärke* 38(7): 224-226.

HOLTEKJØLEN, A.K., UHLEN, A.K., BRATHEN, E., SAHLSTRØM, S. & KNUTSEN, S.H., 2006. Content of starch and non-starch polysaccharides in barley varieties of different origin. *Food Chem*. 94: 348-358.

HOME, S.A, STENHOLM, K.B, WILHELMSON, A.A. & AUTIO, K.A., 2001. Properties of Starch and Cell Wall Components and their Effects on Processing. *Proceedings of the 10th Australian Barley Technical Symposium*, 16-20 September 2001, Canberra, ACT, Australia. Retrieved November 4, 2007, from http://www.regional.org.au/au/abts/

HOOVER, R. & VASANTHAN, T., 1994. Effect of heat-moisture treatment on the structure and physicochemical properties of cereal, legume, and tuber starches. *Carbohydr. Res*. 252: 33-53.

HOOVER, R. & ZHOU, Y., 2003. In vitro and in vivo hydrolysis of legume starches by a-amylase and resistant starch formation in legumes-a review. *Carbohydr. Polym.* 54: 401-417.

HOVENKAMP-HERMELINK, J.H.M., JACOBSEN, E., PONSTEIN, A.S., VISSER, R.G.F., VOS-SCHEPERKEUTER, G.H., BIJMOLT, E.W., DE VRIES, J.N., WITHOLT, B. & FEENSTRA, W.J., 1987. Isolation of amylose-free starch mutant of the potato (*Solanum tuberosum* L.). *Theor. Appl. Genet*. 75: 217-221.

HUBER, K.C. & BEMILLER J.N., 1997. Visualisation of channels and cavities of corn and sorghum starch granules. *Cereal Chem*. 74: 537-541.

HUBER, K.C. & BEMILLER, J.N., 2000. Channels of maize and sorghum starch granules. *Carbohydr. Polym.* 41: 269-276.

HUCL, P. & CHIBBAR, R.N., 1996. Variation for Starch Concentration in Spring Wheat and Its Repeatability Relative to Protein Concentration. *Cereal Chem*. 73(6): 756-758.

HUNG, P.V., MAEDA, T. & MORITA, N., 2006. Waxy and high-amylose wheat starches and flours-characteristics, functionality and application. *Trends Food Sci. Technol.* 17: 448-456.

HUNG, P.V., MAEDA, T., MISKELLY, D., TSUMORI, R. & MORITA, N., 2008. Physicochemical characteristics and fine structure of high-amylose wheat starches isolated from Australian wheat cultivars. *Carbohydr. Polym.* 71(4): 656-663.

HUNG, P.V. & MORITA, N. 2005. Physicochemical properties of hydroxypropylated and cross-linked starches from A-type and B-type wheat starch granules. *Carbohydr. Polym*. 59: 239-246.

HUNG, P.V., YAMAMORI, M. & MORITA, N., 2005. Formation of enzymeresistant starch in bread as affected by high-amylose wheat flour substitutions. *Cereal Chem.* 82: 690-694.

HURKMAN, W.J., MCCUE, K.F., ALTENBACH, S.B., KORN, A., TANAKA, C.K., KOTHARI, K.M., JOHNSON, E.L., BECHTEL, D.B., WILSON, J.D., ANDERSON, O.D. & DUPONT, F.M., 2003. Effect of temperature on expression of genes encoding enzymes for starch biosynthesis in developing wheat endosperm. *Plant Sci*. 164: 873-881.

HYLTON, C.M., DENYER, K., KEELING, P.L., CHANG, M.-T. & SMITH, A.M., 1996. The effect of waxy mutations of the granule-bound starch synthases of barley and maize endosperms. *Planta* 198: 230-237.

HYLTON, C. & SMITH, A.M., 1992. The *rb* mutation of peas caused structural and regulatory changes in ADP glucose pyrophosphorylase from developing embryos. *Plant Physiol*. 99: 1626-1634.

IDF, 2006. Comprehensive review of scientific literature pertaining to nitrogen protein conversion factors. *Bulletin of the International Dairy Federation* No. 405/2006. ISSN 0250-5118, 11 p.

IMBERTY, A., BULEON, A., TRAN, V. & PEREZ, S., 1991. Recent advances in knowledge of starch structure. *Starch/Stärke* 43: 375-384.

IMBERTY, A., CHANZY, H., PEREZ, S., BULEON, A. & TRAN, V., 1988. A revisit to the three-dimensional structure of B-type starch. *Journal of Molecular Biology* 201: 365-378.

IMBERTY, A. & PEREZ, S., 1988. The double-helical nature of the crystalline part of A-starch. *Biopolymers* 27: 1205-1221.

INGLEDEW, W.M., 1993. Yeasts for production of fuel ethanol. In: Rose, A.H., Harrison, J.S. (Eds.). *The yeasts, vol. 5, yeast technology*. New York: Academic Press, pp. 245-291.

INGLEDEW, W.M., 2005. Improvements in Alcohol Technology Through Advancements in Fermentation Technology. *1st European Bioethanol Technology Meeting*. Retrieved November 4, 2007, from http://www.agfdt.de/loads/bi05/ingleimp.pdf

INOUCHI, N., HIBIU, H., LI, T., HORIBATA, T., FUWA, H. & ITANI, T., 2005. Structure and properties of endosperm starch from cultivated rice from Asia and other countries. *J. Appl Glycosci*. 52: 239-246.

INOUCHI, N., GLOVER, D.V. & FUWA, H., 1987. Chain length distribution of amylopectins of several single mutants and the normal counterpart, and sugary-1 phytoglycogen in maize. *Starch/Stärke* 39: 259-265.

ISAKSSON, T., MILLER, C.E. & NÆS, T., 1992. Non-destructive NIR and NIT Determination of Protein, Fat and Water in Plastic-wrapped, Homogenized Meat. *Appl. Spectrosc*. 46(11): 1685.

ISHADA, N., MIURA, H., NODA, T. & YAMAUCHI, H., 2003. Physical properties of white salted noodles from near-isogenic wheat lines with different wax protein-deficiency. *Starch/Stärke* 55: 390-396.

ITO, T., SAITO, K., SUGAWARA, M., MOCHIDA, K. & NAKAKUKI, T., 1999. Effect of raw and heat-moisture-treated highamylose corn starches on the process of digestion in the rat digestive tract. *J. Sci. Food Agric*. 79: 1203-1207.

JACOBI, A. & HARTMANN, F., 2005. Breeding of cereal crops for the production of bioethanol. *1st European Bioethanol Technology Meeting*. Retrieved November 4, 2007, from http://www.agfdt.de/loads/bi05/jacobi.pdf

JACOBSEN, S., SONDERGAARD, I., MOLLER, B., DESLER, T. & MUNCK, L., 2005. A chemometric evaluation of the underlying physical and chemical patterns that support near infrared spectroscopy of barley seeds as a tool for explorative classification of endosperm genes and gene combinations. *J. Cereal Sci.* 42: 281-299.

JAMES, M.G., DENYER, K. & MYERS, A.M., 2003. Starch synthesis in the cereal endosperm. *Curr. Opin. Plant Biol*. 6: 215-222.

JAMES, M.G., ROBERTSON, D.S. & MYERS, A.M., 1995. Characterization of the maize gene sugary 1, a determinant of starch composition in kernels. *Plant Cell.* 7: 417-429.

JANE, J., 2006. Current Understanding on Starch Granule Structures. *J. Appl. Glycosci*. 53: 205-213.

JANE, J., AO, Z., DUVICK, S.A., YOO, S.-H., WONG, K.S. & GARDNER, C., 2003. Structures of amylopectin and starch granules: How are they synthesized? *J. Appl. Glycosci.* 50: 167-171.

JANE, J. & CHEN, J.F., 1992. Effect of amylose molecular size and amylopectin branch chain length on paste properties of starch. *Cereal Chem*. 69: 60-65.

JANE, J., CHEN, Y.Y., LEE, L.F., MCPHERSON, A.E., WONG, K.S., RADOSAVLJEVIC, M. & KASEMSUWAN, T., 1999. Effects of amylopectin branch chain length and amylose content on the gelatinization and paste properties of starch. *Cereal Chem.* 76: 629-637.

JANE, J., KASEMSUWAN, T., LEAS, S., ZOBEL, H. & ROBYT, J.F., 1994. Anthology of starch granule morphology by scanning electron microscopy. *Starch/Stärke* 46: 121-129.

JANE, J.L. & SHEN, J.J., 1993. Internal structure of the potato starch granule revealed by chemical gelatinisation. *Carbohydr. Res*. 247: 279-290.

JANE, J., SHEN, L., WANG, L. & MANINGAT, C.C., 1992. Preparation and Properties of Small-Particle Corn Starch. *Cereal Chem*. 69: 280-283.

JANE, J.L., XU, A., RADOSAVLJEVIC, M. & SEIB, P.A., 1992. Location of amylase in normal starch granules. I. Susceptibility of amylase and amylopectin in cross-linking reagents. *Cereal Chem*. 69: 405-409.

JENKINS, P.J., CAMERON, R.E. & DONALD, A.M., 1993. A universal feature in the structure of starch granules from different botanical sources. *Starch/Stärke* 45: 417-420.

JENKINS, P.J. & DONALD, A.M., 1994. The influence of amylose on starch granule structure. *Int. J. Biol Macromol*. 17: 315-321.

JENNER, H., 2003. Transgenesis and yield: what are our targets? *Trends Biotechnol*. 21: 190-192.

JENNER, C.F., DENYER, K. & GUERIN, J., 1995. Thermal characteristics of soluble starch synthase from wheat endosperm. *Australian Journal of Plant Physiology* 22: 703-709.

JESPERSEN, H.M., MACGREGOR, E.A., HENRISSAT, B., SIERKS, M.R. & SVENSSON, B., 1993. Starch- and glycogen-debranching and branching enzymes: prediction of structural features of the catalytic $(b/a)_8$-barrel domain and evolutionary of other amylolytic enzymes. *J. Prot. Chem*. 12: 791-805.

JIANG, D., TAO, Q.N. & CAO, W.X., 2002. Effects of water logging on accumulation and redistribution of water soluble carbohydrate metabolism in culm of wheat. *Acta Agron. Sin.* 28: 230-234.

JING, Z.P., GALLARDO, F., PASCUAL, M.B., SAMPALO, R., ROMERO, J., TORRES DE NAVARRA, A. & CANOVAS, F.M., 2004. Improved growth in a field trial of transgenic hybrid poplar overexpressing glutamine synthetase. *New Phytol.* 164: 137-145.

JOBLING, S., 2004. Improving starch for food and industrial applications. *Current Opinion in Plant Biology* 7: 210-218.

JOBLING, S.A., JARMAN, C., THE, M.M., HOLMBERG, N., BLAKE, C. & VERHOEYEN, M.E., 2003. Immunomodulation of enzyme function in plants by single-domain antibody fragments. *Nat. Biotechnol*. 21: 77-80.

JOBLING, S.A., SCHWALL, G.P., WESTCOTT, R.J., SIDEBOTTOM, C.M., DEBET, M., GIDLEY, M.J., JEFFCOAT, R. & SAFFORD, R., 1999. A minor form of starch branching enzyme in potato (*Solanum tuberosum* L.) tubers has a major effect on starch structure: cloning and characterization of multiple forms of SBE A. *Plant J.* 18: 163-171.

JOHNSON, P.E., PATRON, N.J., BOTTRILL, A.R., DINGES, J.R., FAHY, B.F., PARKER, M.L., WAITE, D.N. & DENYER, K., 2003. A low-starch barley mutant, Riso 16, lacking the cytosolic small subunit of ADP-glucose pyrophosphorylase, reveals the importance of the cytosolic isoform and the identity of the plastidial small subunit. *Plant Physiol*. 131: 684-696.

JUSZCZAK, L., 2003. Surface of triticale starch granules – NC-AFM observations. *Electronic Journal of Polish Agricultural Universities, Food Science and Technology* 6(1). Retrieved November 4, 2007, from http://www.ejpau.media.pl/series/volume6/issue1/food/art-08.html

JUSZCZAK, L., FORTUNA, T. & KROK, F., 2003. Non-contact Atomic Force Microscopy of starch granules surface. Part II. Selected cereal starches. *Starch/Stärke* 55(1): 8-16.

KAIZUMA, K., 1988. Structure and chemistry of the starch granule. In: J. Preiss (Ed.), *The Biochemistry of Plants*, Vol. 14. Carbohydrates. San Diego: Academic Press, pp. 141-180.

KALUGINA, N.A. & ZAPOL'SKAYA, E.B., 2001. Biologicheski razrushaemye materialy na osnove krakhmala (in Russian). In: *Krakhmal i krakhmalsoderzhashchie istochniki – struktura, svojstva i novye tekhnologii:* Sbornik dokladov mezhdunarodnoj konferentsii. Moscow, p. 132.

KARADJ, A.M., STODDARD, F.L. & MARSHALL, D.R., 1999. Amylose content in segregating populations of einkorn, emmer, and rye. *Starch/Stärke* 51(2-3): 66-73.

KARPENSTEIN-MACHAN, M. & SCHEFFER, K., 1998. Triticale for industrial uses, produced in a sustainable cropping system. In: P. Juskiw (Ed.), *Proceedings of the 4th International Triticale Symposium*, Red Deer, Alberta, Canada, Vol. I, pp. 273-277.

KATOPO, H., SONG, Y. & JANE, J.-L., 2002. Effect and mechanism of ultrahigh hydrostatic pressure on the structure and properties of starches. *Carbohydr. Polym.* 47: 233-244.

KATZ, F.R., FURCSIK, S.L., TENBARGE, F.L., HAUBER, R.J. & FRIEDMAN, R.B., 1993. Behavior of starches derived from varieties of maize containing different genetic mutations: effects of starch genotype on granular morphology. *Carbohydr. Polym*. 21(2): 133-136.

KAYS, S.E., BARTON II, F.E. & WINDHAM, W.R., 2000. Predicting protein content by near infrared reflectance spectroscopy in diverse cereal food products. *J. Near Infrared Spectrosc*. 8: 35-43.

KEELING, P.L., BACON, P.J. & HOLT, D.C., 1993. Elevated Temperature Reduces Starch Deposition in Wheat Endosperm by Reducing the Activity of Soluble Starch Synthase. *Planta* 191: 342-348.

KEELING, P.L., WOOD, J.R., TYSON, R.H. & BRIDGES, I.G., 1988. Starch biosynthesis in developing wheat grain. *Plant Physiol.* 87: 311-19.

KELSALL, D.R. & LYONS, T.P., 2003. Practical management of yeast: conversion of sugars to ethanol. In: K.A. Jaques, T.P. Lyons, D.R. Kelsall (Eds.), *The Alcohol Textbook*. Nottingham University Press, Nottingham, UK.

KEYZER, M.A., MERBIS, M.D. & VOORTMAN, R.L., 2008. The Biofuel Controversy. *De Economist* 156: 507-527.

KIM, S. & DALE, B.E., 2004. Global potential bioethanol production from wasted crops and crop residues. *Biomass and Bioenergy* 26: 361-375.

KIM, K.-N., FISHER, D.K., GAO, M. & GUITINAN, M.J., 1998. Molecular cloning and characterization of the amylose extender gene encoding starch branching enzyme IIb. *Plant Mol. Biol.* 38: 945-956.

KIM, W., JOHNSON, J.W., GRAYBOSCH, R.A. & GAINES, C.S., 2003. Physicochemical properties and end-use quality of wheat starch as a function of waxy protein alleles. *J. Cereal Sci*. 37: 195-204.

KIMURA, A. & ROBYT, J.F., 1995. Reaction of enzyme with starch granules: kinetics and products of the reaction with glucoamylase. *Carbohydr. Res*. 277: 87-107.

KINDRED, D.R., SMITH, T.C., SYLVESTER-BRADLEY, R., GINSBERG, D. & DYER, C.J., 2007. Optimising nitrogen applications for wheat grown for the biofuels market. *HGCA Project Report No. 417*, 44 pp.

KINDRED, D.R., VERHOEVEN, T.M.O., WEIGHTMAN, R.M., SWANSTON, J.S., AGU, R.C., BROSNAN, J.M. & SYLVESTER-BRADLEY, R., 2008. Effects of variety and fertiliser nitrogen on alcohol yield, grain yield, starch and protein content, and protein composition of winter wheat. *J. Cereal Sci.* 48: 46-57.

KIRIBUCHI-OTOBE, C., NAGAMINE, T., YANAGISAWA, T., OHNISH, M. & YAMAGUCHI, I. 1997. Production of hexaploid wheats with waxy endosperm character. *Cereal Chem.* 74: 72-74.

KISELEVA, V.J., KRIVANDIN, A.V., FORNAL, J., BLASZCZAK, W., JELINSKI, T. & YURYEV, V.P., 2005. Annealing of normal and mutant wheat starches. LM, SEM, DSC, and SAXS studies. *Carbohydrate Res.* 340: 75-83.

KISHIDA, T., NOGAMI, H., HIMENO, S. & EBIHARA, K., 2001. Heat moisture treatment of high amylose corn starch increases its resistant starch content but not its physiological effects in rats. *J. Nutr*. 131: 2716-2721.

KLECZKOWSKI, L.A., 2001. A new player in the starch field. *Plant Physiol. Biochem*. 39: 759-761.

KLESCHENKO, A.D., ZOIDZE, E.K. & BOKEN, V.K., 2005. Monitoring agricultural drought in Russia. In: Boken, V.K., Cracknell, A.P. & Heathcote, R.L. (Eds.), *Monitoring and predicting agricultural drought.* World Meteorological Organization, Oxford University Press US, pp. 196-207.

KLOSGREN, R.B., GIERL, A., SCHWARZ-SOMMER, Z. & SAEDLER, H., 1986. Molecular analysis of the waxy locus of *Zea mays*. *Mol. Genet*. 203: 237-244.

KLUCINEC, J. & THOMPSON, D.B., 1999. Amylose and amylopectin interact in the gelation of dispersed high-amylose starches. *Cereal Chem*. 76: 282-291.

KNIGHT, M.E., HARN, C., LILLEY, C.E.R., GUAN, H., SINGLETARY, G.W., MU-FORSTER, C., WASSERMAN, B.P. & KEELING, P.L., 1998. Molecular cloning of starch synthase I from maize (W64) endosperm and expression in *Esherichia coli*. *Plant J.* 14: 613-622.

KNUTSON, C.A. & GROVE, M.J., 1994. Rapid method for estimation of amylose in maize starches. *Cereal Chem*. 71(5): 469-471.

KNUTSON, C.A., KHOO, U., CLUCKEY, J.E. & INGLETT, G.E., 1982. Variation in enzyme digestibility and gelatinization behavior of corn starch granule fractions. *Cereal Chem*. 59(5): 512-515.

KOHYAMA, K. & SASAKI, T., 2006. Differential scanning calorimetry and a model calculation of starches annealed at 20 and 50°C. *Carbohydr. Polym.* 63: 82-88.

KOLIATSOU, M. & PALMER, G.H., 2003. A new method to assess mealiness and steeliness of barley varieties and relationship of mealiness with malting parameters. *Journal of American Society of Brewing Chemists* 61: 114-118.

KOLIATSOU, M. & PALMER, G.H., 2004. Relationship between flour turbidity, malting and endosperm structure. *J. Inst. Brew*. 110: 193-199.

KOROTEEVA, D.A., KISELEVA, V.I., SRIROTH, K., PIYACHOMKWAN, K., BERTOFT, E., YURYEV, P.V. & YURYEV, V.P., 2007. Structural and thermodynamic properties of rice starches with different genetic background. Part 1. Differentiation of amylopectin and amylose defects. *Intl. J. Biol. Macromol.* 41: 391-403.

KOSAR-HASHEMI, B., LI, Z., LARROQUE, O., REGINA, A., YAMAMORI, M., MORELL, M.K. & RAHMAN, S., 2007. Multiple effects of the starch synthase II mutation in developing wheat endosperm. *Functional Plant Biology* 34: 431-438.

KOSARIC, N. & VARDAR-SUKAN, F., 2001. Potential source of energy and chemical products. In: Roehr, M. (Ed.). *The biotechnology of ethanol: classical and future applications.* Wiley-VCH: New York, p. 92.

KOSSMAN, J., ABEL, G.J.W., SPRINGER, F., LLOYD, J.R. & WILLIMIZER, L., 1999. Cloning and functional analysis of a cDNA encoding a starch synthase from potato (*Solanum tuberosum* L.) that is predominantly expressed in leaf tissue. *Planta* 208: 503-511.

KOSSMAN, J. & LLOYD, J., 2000. Understanding and influencing starch biochemistry. *Critical Rev. in Plant Sci*. 19: 171-226.

KOUTINAS, A.A., WANG, R. & WEBB, C., 2004. Evaluation of wheat as generic feedstock for chemical production. *Industrial Crops and Products* 20: 75-88.

KOVALENKO, I.V., RIPPKE, G.R. & HURBURGH, C.R., 2006. Determination of amino acid composition of soybeans (*Glycine max*) by near-infrared spectroscopy. *J. Agric. Food Chem.* 54: 3485-3491.

KOZLOV, S.S., BLENNOW, A., KRIVANDIN, A.V. & YURYEV, V.P., 2007a. Structural and thermodynamic properties of starches extracted from GBSS- and GWD suppressed potato lines. *Intl. J. Biol. Macromol.* 40: 449-460.

KOZLOV, S.S., KRIVANDIN, A.V., SHATALOVA, O.V., NODA, T., BERTOFT, E., FORNAL, J. & YURYEV, V.P., 2007b. Structure of starches extracted from near-isogenic wheat lines: Part 2. Molecular organization of amylopectin clusters. *J. Thermal Analysis and Calorim.* 87(2): 575-584.

KOZLOV, S.S., NODA, T., BERTOFT, E. & YURYEV, V.P., 2006. Structure of starches extracted from near isogenic wheat lines. Part I. Effect of different GBSS I combinations. *J. Thermal Analysis and Calorim.* 86(2): 291-301.

KRETOVICH, V.L., 1986. Biokhimiya rastenij (in Russian). 2-e izd., Moscow, 504 pp.

KU, M., CHO, D., LI, X., JIAO, D., PINTO, M., MIYAO, M. & MATSUOKA, M., 2001. Introduction of genes encoding C4 photosynthesis enzymes into rice plants: physiological consequences. *Novartis Found Symp.* 236: 100-111; discussion 111-116.

KUBO, A., RAHMAN, S., UTSUMI, Y., LI, Z.Y., MUKAI, Y., YAMAMOTO, M., UGAKI, M., HARADA, K., SATOH, H., KONIK-ROSE, C., MORELL, M. & NAKAMURA, Y., 2005. Complementation of sugary-1 phenotype in rice endosperm with the wheat isoamylase1 gene in supports a direct role for isoamylase1 amylopectin biosynthesis. *Plant Physiol.* 137: 43-56.

KUBO, A., YUGUCHI, Y., TAKEMASA, M., SUZUKI, S., SATOH, H. & KITAMURA, S., 2008. The use of micro-beam X-ray diffraction for the characterization of starch crystal structure in rice mutant kernels of *waxy*, *amylose extender*, and *sugary1*. *J. Cereal Sci.* 48(1): 92-97.

KUCEROVA, J., 2006. Usability of triticale into bioethanol producing through the use of technical enzymes / Vhodnost tritikale k vyrobe bioethanolu pri pouziti technickych enzymu. *Acta Universitatis Agriculturae et Silviculturae Mendelianae Brunensis* 54(4): 33-38.

KUIPERS, A.G.J., JACOBSEN, E. & VISSER, R.G.F., 1994. Formation and deposition of amylose in the potato tuber are affected by the reduction of granule-bound starch synthase gene expression. *Plant Cell*. 6: 43-52.

KUMAR, I. & KHUSH, G.S., 1986a. Genetics of amylose content in rice (*Oryza sativa* L.). *J. Genet.* 65: 1-11.

KUMAR, I. & KHUSH, G.S., 1986b. Gene dosage effects of amylose content in rice endosperm. *Japanese J. Genetics* 61: 559-568.

KUMAR, I. & KHUSH, G.S., 1988. Inheritance of amylose content in rice (*Oryza sativa* L.). *Euphytica* 38: 261-269.

LANGEVELD, S.M.J., VAN WIJK, R., STUURMAN, N., KIJNE, J.W. & DE PATER, S., 2000. B-type granule containing protrusions and interconnections between amyloplasts in developing wheat endosperm revealed by transmission electron microscopy and GFP expression. *J. Exp. Bot*. 51: 1357-1361.

LARSON, B., 2004. Crop Management. Hybrid Characterization Test for Ethanol Yields Results. *Syngenta Seeds*. Posted 10 August 2004. Retrieved November 4, 2007, from http://www.plantmanagementnetwork.org/pub/cm/news/2004/ethanol/

LASZTITY, R., 1984. Rye and triticale proteins. In: *The Chemistry of Cereal Proteins*. Chapter 4. CRC Press, Florida, pp. 103-115.

LAURO, M., FORSSELL, P.M., SUORTTI, M.T., HULLEMAN, S.H.D. & POUTENEN, K.S., 1999. Alpha-amylolysis of large barley starch granules. *Cereal Chem.* 7(6): 925-930.

LEA, P.J. & AZEVEDO, R.A., 2007. Nitrogen use efficiency. 2. Amino acid metabolism. *Annals of Applied Biology* 151: 269-275.

LEE, M.R., SWANSON, B.G. & BAIK, B.K., 2001. Influence of amylose content on properties of wheat starch and breadmaking quality of starch and gluten blends. *Cereal Chem*. 78: 701-706.

LEGGEWIE, G., KOLBE, A., LEMOINE, R., ROESSNER, U., LYTOVCHENKO, A., ZUTHER, E., KEHR, J., FROMMER, W., RIESMEIER, J., WILLMITZER, L. & FERNIE, A., 2003. Overexpression of the sucrose transporter SoSUT1 in potato results in alterations in leaf carbon partitioning and in tuber metabolism but has little impact on tuber morphology. *Planta* 217: 158-167.

LEVINE, L.H., BAUER, J. & LEVINE, H.G., 2005. Critical Aspects of Starch Determination in Plant Tissues and a New Approach Utilizing HPAEC/PAD for the Quantification of Starch-Derived Glucose. *SAE Technical Papers* 2005-01-2773.

LI, L., 2006. Starch biogenesis: relationship between starch structures and starch biosynthetic enzymes. Ph.D. Dissertation. Iowa State University, Ames, LA, USA.

LI, Z., CHU, X., MOUILLE, G., YAN, L., KOSAR-HASHEMI, B., HEY, S., NAPIER, J., SHEWRY, P., CLARKE, B., APPELS, R., MORELL, M.K. & RAHMAN, S., 1999. The localization and expression of the class II starch synthases of wheat. *Plant Physiol.* 120: 1147-1156.

LI, Z., HUANG, B., RAMPLING, L., WANG, J., YU, J., MORELL, M. & RAHMAN, S., 2004. Detailed comparison between the wheat chromosome group 7 short arms and the rice chromosome arms 6S and 8L with special reference to genes involved in starch biosynthesis. *Funct. Integr. Genomics* 4: 231-240.

LI, J.H., VASANTHAN, T., HOOVER, R. & ROSSNAGEL, B.G., 2004. Starch from hull-less barley. V. In vitro susceptibility of waxy, normal, and high-amylose starches towards hydrolysis by alpha-amylases and amyloglucosidase. *Food Chem*. 84: 621-632.

LIANG, J.S., ZHANG, J.H. & CAO, X.Z., 2001. Grain sink strength may be related to the poor grain filling of indica-japonica rice (*Oryza sativa*) hybrids. *Physiol. Plant*. 112(4): 470-477.

LII, C.-Y., TSAI, M.-L. & TSENG, K.-H., 1996. Effect of Amylose Content on the Rheological Property of Rice Starch. *Cereal Chem*. 73(4): 415-420.

LIM, S.T., JANE, J.L., RAJAGOPALAN, S. & SEIB, P.A., 1992. Effects of Starch Granule Size on Physical Properties of Starch-Filled Polyethylene Film. *Biotechnol. Prog*. 8: 51-55.

LINEBACK, D.R. & RASPER, V.F., 1998. Wheat carbohydrates. In: Y. Pomeranz (Ed.), *Wheat Chemistry and Technology*, Volume 1. American Association of Cereal Chemists, Minnesota, USA, p. 281.

LIU, Q. & THOMPSON, D.B., 1998. Effects of moisture content and different gelatinization heating temperature on retrogradation of waxy-type maize starches. *Carbohydr. Res*. 314: 221-235.

LLOYD, J.R., SPRINGER, F., BULEON, A., MULLER-ROBER, B., WILLMITZER, L. & KOSSMANN, J., 1999. The influence of alterations in ADP-glucose pyrophosphorylase activities on starch structure and composition in potato tubers. *Planta* 209: 230-238.

LORBERTH, R., RITTE, G., WILLMITZER, L. & KOSSMANN, J., 1998. Inhibition of a starch-granule-bound protein leads to modified starch and repression of cold sweetening. *Nat. Biotechnol*. 16: 473-477.

LOYCE, C. & MEYNARD, J.M., 1997. Low input wheat management techniques are more efficient in ethanol production. *Industr. Crops Prod.* 6: 271-283.

LOYCE, C., RELLIER, J.P. & MEYNARD, J.M., 2002. Management planning for winter wheat with multiple objectives (2): ethanol-wheat production. *Agricultural Systems* 72: 33-57.

LU, G., HUANG, H. & ZHANG, D., 2006. Prediction of sweetpotato starch physiochemical quality and pasting properties using near-infrared reflectance spectroscopy. *Food Chem.* 94: 632-639.

LU, G.Q. & SHENG, J.L., 1990. Application of near infrared reflectance spectroscopy (NIRS) in sweet potato quality breeding. *Scientia Agricultura Sinica* 23: 76-81.

LU, T.-J., PAI, Y.-Y. & LU, C.-Y., 1999. The susceptibility of A- and B-type wheat starch granules to bacterial alpha-amylase. Abstract 241. *Cereal Foods World* 44: 545.

LUSTINEC, J., HADACOVA, V., KAMINEK, M. & PROCHAZKA, Z., 1983. Quantitative determination of starch, amylose, and amylopectin in plant tissues using glass fiber paper. *Anal Biochem*. 132(2): 265-271.

MABEE, W.E., SADDLER, J.N., NIELSEN, C., NIELSEN, L.H. & JENSEN, E.S., 2006. Renewable-based fuels for transport. In: *Renewable energy for power and transport*. Risø energy report 5, November 2006, p. 47-50.

MACLEOD, L.C. & DUFFUS, C.M., 1988. Reduced Starch Content and Sucrose Synthase Activity in Developing Endosperm of Barley Plants Grown at Elevated Temperatures. *Aust. J. Plant Physiol*. 15: 367-75.

MANGALIKA, W.H.A., MIURA, H., YAMAUCHI, H. & NODA, T., 2003. Properties of starches from near-isogenic wheat lines with different Wx protein deficiencies. *Cereal Chem.* 80(6): 662-666.

MANNERS, D.J., 1989. Recent developments in our understanding of amylopectin structure. *Carbohydr. Polym*. 11(1): 87-112.

MARAIS, G.F., BOTES, W.C. & LOUW, J.H., 2001. Wheat breeding based on recurrent mass selection. *Cereal Res. Comm*. 29: 339-342.

MARK, H., 2001. Fundamentals of near-infrared spectroscopy. In: Raghavachan, R. (Ed.), *Near-infrared Applications in Biotechnology*. Marcel Dekker, New York, pp. 293-321.

MARTENS, H. & NÆS, T., 1989. Multivariate Calibration. Wiley, Chichester, UK, 419 pp.

MARTENS, H. & NÆS, T., 1991. Multivariate Calibration. Wiley, Chichester, UK, p. 7, 345.

MARTENS, H. & NÆS, T., 2001. Multivariate calibration by data compression. In: Williams P., Norris K. (Eds.), *Near-Infrared Technology in the Agricultural and Food Industries*, Chapter 4. American Association of Cereal Chemists, Inc. St. Paul, MN, pp. 59-100.

MARTIN, C. & SMITH, A.M., 1995. Starch biosynthesis. *Plant Cell*. 7: 971-985.

MARTINANT, J.-P., BILLOT, A., BOUGUENNEC, A., CHARMET, G. & SAULNIER, L.G.B., 1999. Genetic and environmental variations in water extractable arabinoxylans content and flour extract viscosity. *J. Cereal Sci.* 30: 45-48.

MARTINANT, J.-P., CADALEN, T., BILLOT, A., CHARTIER, S., LEROY, P., BERNARD, M. & SAULNIER, L.G.B., 1998. Genetic analysis of water-extractable arabinoxylans in bread wheat endosperm. *Theor. Appl. Genet.* 97: 1069-1075.

MARTINEZ, C. & PRODOLLIET, J., 1996. Determination of Amylose in Cereal and Non-Cereal Starches by a Colorimetric Assay: Collaborative Study. *Starch/Stärke* 48(3): 81-85.

MATS'KOVYAK, V., 1990. Dostizheniya pol'skoj selektsii triticale po dannym natsional'nykh i mezhdunarodnykh rezul'tatov (in Russian). *Sb. nauch. trud. koordinatsionnogo soveshchaniya stran-chlenov SEV*, 25-29 iyunya 1990 g. ISAR, Radzikov, Pol'sha, pp. 3-17.

MATSUOKA, M., FURBANK, R., FUKAYAMA, H. & MIYAO, M., 2001. Molecular engineering of C4 photosynthesis. *Annu. Rev. Plant Physiol. Plant Mol. Biol*. 52: 297-314.

MCCLEARY, B.V., GIBSON, T.S. & MUGFORD, D.C., 1997. Measurement of total starch in cereal products by amyloglucosidase – α-amylase method: Collaborative study. *Journal of the Association of Official Analytical Chemists* 80: 571-579.

MCCLEARY, B.V., SOLAH, V. & GIBSON, T.S., 1994. Quantitative Measurement of Total Starch in Cereal Flours and Products. *J. Cereal Sci*. 20: 51-58.

MCGRANCE, S.J., CORNELL, H.J. & RIX, C.J., 1998. A Simple and Rapid Colorimetric Method for the Determination of Amylose in Starch Products. *Starch/Stärke* 50(4): 158-163.

MCKENZIE, K.S. & RUTGER, J.N., 1983. Genetic analysis of amylose content, alkali spreading score, and grain dimensions in rice. *Crop Sci.* 23: 306-313.

MCLAREN, J.S., 2005. Crop biotechnology provides an opportunity to develop a sustainable future. *Trends in Biotech.* 23: 339-342.

MCLEOD, J.G., BARON, V.S., SALMON, D.F. & GAN, Y.T., 1998. Triticale – potential as an industrial ethanol feedstock. In: Juskiw, P. (Ed.), *Proceedings of the 4th International Triticale Symposium*, Vol. I. Red Deer, Alberta, Canada, pp. 278-283.

MCLEOD, J.G., GAN, Y.T. & BARON, V.S., 1997. An assessment of the cereal resources of Western Canada for ethanol feedstock. In: Stumborg, M.A. (Ed.), *Proc. 1997 Ethanol research and development workshop*. Pub. NRC/AAFC, pp. 5-8.

MEREDITH, P., 1981. Large and small granules in wheat – are they really different? *Starch/Stärke* 33: 40-44.

MESTRES, C., MATENCIO, F., PONS, B., YAJID, M. & FLIEDEL, G., 1996. A rapid method for the determination of amylose content by using differential scanning calorimetry. *Starch/Stärke* 48: 2-6.

MEYER, F.D., SMIDANSKY, E.D., BEECHER, B., GREENE, T.W. & GIROUX, M.J., 2004. The maize Sh2r6hs ADP-glucose pyrophosphorylase (AGP) large subunit confers enhanced AGP properties in transgenic wheat (*Triticum aestivum*). *Plant Sci.* 167: 899-911.

MIFLIN, B. & HABASH, D., 2002. The role of glutamine synthetase and glutamate dehydrogenase in nitrogen assimilation and possibilities for improvement in the nitrogen utilization of crops. *J. Exp. Bot.* 53: 979-987.

MIKU, V.E., 1981. Geneticheskie issledovaniya kukuruzy (in Russian). Kishinev, Shtiintsa, 231 pp.

MILLAR, S.J., 2003. The development of near-infrared (NIR) spectroscopy calibrations for the prediction of wheat and flour quality. *The Home Grown Cereals Authority (HGCA) Project Report No. 310*. HGCA: London.

MILLER, C.E., 2001. Chemical principles in near-infrared technology. In: Williams, P., Norris, K. (Eds.), *Near Infrared Technology in Agricultural and Food Industries*. American Association of Cereal Chemists, St. Paul, Minnesota. USA, pp. 19-38.

MIRALBES, C., 2004. Quality control in the milling industry using near infrared transmittance spectroscopy. *Food Chem.* 88: 621-628.

MIURA, H., ARAKI, E. & TANII, S., 1999. Amylose synthesis capacity of the three Wx genes of wheat cv. Chinese Spring. *Euphytica* 108: 91-95.

MIURA, H. & SUGAWARA, A., 1996. Dosage effects of the three *Wx* genes on amylose synthesis in wheat endosperm. *Theor. Appl. Genet*. 93: 1066-1070.

MIURA, H. & TANII, S., 1994. Endosperm starch properties in several wheat cultivars preferred for Japanese noodles. *Euphytica* 72: 171-175.

MIURA, H., TANII, S., NAKAMURA, T. & WATANABE, N., 1994. Genetic control of amylose content in wheat endosperm starch and differential effects of three Wx genes. *Theor. Appl. Genet*. 89: 276-280.

MIURA, H., WICKRAMASINGHE, M.H.A., SUBASINGHE, R.M., ARAKI, E. & KOMAE, K., 2002. Development of near-isogenic lines of wheat carrying different null *Wx* alleles and their starch properties. *Euphytica* 123: 353-359.

MIZUNO, K., KAWASAKI, T., SHIMADA, H., SATOH, H., KOBAYASHI, E., OKUMURA, S., ARAI, Y. & BABA, T., 1993. Alteration of the structural properties of starch components by the lack of an isoform of starch branching enzyme in rice seeds. *J. Biol. Chem.* 268: 19084-19091.

MIZUNO, K., KIMURA, K., ARAI, Y., KAWASAKI, T., SHIMADA, H. & BABA, T., 1992. Starch branching enzymes from immature rice seeds. *J. Biochem*. 112: 643-651.

MIZUNO, K., KOBAYASHI, E., TACHIBANA, M., KAWASAKI, T., FUJIMURA, T., FUNANE, K., KOBAYASHI, M. & BABA, T., 2001. Characterization of an isoform of rice starch branching enzyme, RBE4, in developing seeds. *Plant Cell Physiol.* 42: 349-357.

MOHAMMADKHANI, A., 2005. Survey of Starch Amylose Content in Naked Barley (*H. vulgare Nudum*). *Pakistan Journal of Nutrition* 4(3): 183-186.

MOHAMMADKHANI, A., STODDARD, F.L. & MARSHALL, D.R., 1998. Survey of amylose content in *Secale cereale, Triticum monococcum, T. turgidum* and *T. tauschii*. *J. Cereal Sci*. 28: 273-280.

MOHAMMADKHANI, A., STODDARD, F.L. & MARSHALL, D.R., 1999a. Amylose Content in Segregating Populations of Einkorn, Emmer, and Rye. *Starch/Stärke* 51(2-3): 66-73.

MOHAMMADKHANI, A., STODDARD, F.L., MARSHALL, D.R., UDDIN, M.N. & ZHAO, X., 1999b. Starch Extraction and Amylose Analysis from Half Seeds. *Starch/Stärke* 51(2-3): 62-66.

MOJOVIC, L., NIKOLIC, S., RAKIN, M. & VUKASINOVIC, M., 2006. Production of bioethanol from corn meal hydrolyzates. *Fuel* 85: 1750-1755.

MONSANTO INC., 2007. Processor Preferred®: High Fermentable and High Extractable Corn. Monsanto Inc. Retrieved November 4, 2007, from http://www.monsanto.com/monsanto/ag_products/output_traits/hfc_corn.asp

MOORTHY, S.N., 2002. Physicochemical and functional properties of tropical tuber starches: a review. *Starch/Stärke* 54: 559-592.

MORALES, M.D., ESCARPA, A. & GONZALEZ, M.C., 1997. Simultaneous determination of resistant and digestible starch in foods and food products. *Starch/Stärke* 49(11): 448-453.

MORELL, M.K., BLENNOW, A., KOSAR-HASHEMI, B. & SAMUEL, M.S., 1997. Differential expression and properties of starch-branching enzyme isoforms in developing wheat endosperm. *Plant Physiol*. 113: 201-208.

MORELL, M.K., KONIK-ROSE, C., AHMED, R., LI, Z. & RAHMAN, S., 2004. Synthesis of resistant starches in plants. *J. AOAC Int.* 87: 740-748.

MORELL, M.K., KOSAR-HASHEMI, B., CMIEL, M., SAMUEL, M.S., CHANDLER, P., RAHMAN, S., BULEON, A., BATEY, I.L. & LI, Z., 2003. Barley sex6 mutants lack starch synthase IIa activity and contain a starch with novel properties. *Plant J.* 34(2): 173-185.

MORELL, M.K. & MYERS, A.M., 2005. Towards the rational design of cereal starches. *Curr. Opin. Plant Biol*. 8: 204-210.

MORELL, M.K., RAHMAN, S., ABRAHAMS, S.L. & APPELS, R., 1995. The biochemistry and molecular biology of starch synthesis in cereals. *Aust. J. Plant Physiol*. 22: 647-660.

MORGAN, J.E. & WILLIAMS, P.C., 1995. Starch damage in wheat flours: A comparison of enzymatic, iodometric, and near infrared reflectance techniques. *Cereal Chem.* 72: 209-212.

MORRIS, C.F., GREENBLATT, G.A., BETTGE, A.D. & MALKAWI, H.I., 1994. Isolation and characterization of multiple forms of friabilin. *J. Cereal Sci*. 20: 167-174.

MORRISON, W.R. & GADAN, H.J., 1987. The amylose and lipid contents of starch granules in developing wheat endosperm. *J. Cereal Sci*. 5: 263-275.

MORRISON, W.R., SCOTT, D.C. & KARKALAS, J., 1986. Variation in the composition and physical properties of barley starches. *Starch/Stärke* 38(11): 374-379.

MOSSÉ, J., 1990. Nitrogen to protein conversion factor for ten cereals and six legumes or oilseeds. A reappraisal of its definition and determination. Variation according to species and to seed protein content. *J. Agric. Food Chem*. 38: 18-24.

MOUILLE, G., MADDELEIN, M.-L., LIBESSART, N., TALAGA, P., DECQ, A., DELRUE, B. & BALL, S., 1996. Preamylopectin processing: a mandatory step for starch biosynthesis in plants. *The Plant Cell* 8: 1353-1366.

MU, C., HARN, C., KO, Y.-T., SINGLETARY, G.W., KEELING, P.L. & WASSERMAN, B.P., 1994. Association of a 76Kda polypeptide with soluble starch synthase I activity in maize (cv B73) endosperm. *Plant J.* 6(2): 151-159.

MÜLLER-RÖBER, B. & KOSSMANN, J., 1994. Approaches to influence starch quantity and starch quality in transgenic plants. *Plant Cell Environ.* 17: 601-613.

MÜLLER-RÖBER, B., SONNEWALD, U. & WILLMITZER, L., 1992. Inhibition of the ADP-Glucose Pyrophosphorylase in Transgenic Potatoes Leads to Sugar-Storing Tubers and Influences Tuber Formation and Expression of Tuber Storage Protein Genes. *EMBO J.* 11: 1229-1238.

MULUK, C., 1996. Utilization of near infrared reflectance spectroscopy for food corn hybrid evaluation. Ph.D. abstract, AAT 9713766, University of Kentucky, 210 pp.

MUNCK, L., 2003. Detecting diversity – a new holistic, exploratory approach bridging the geneotype and phenotype. In: von Bothmer, R., van Hintum, T.J.L., Knupffer, H., Sato, K. (Eds.), *Diversity in Barley* (*Hordeum vulgare*). Elsevier Science B.V., Amsterdam, The Netherlands, pp. 227-245.

MUNCK, L. & MØLLER, B., 2004. A new germinative classification model of barley for prediction of malt quality amplified by a near infrared transmission spectroscopy calibration for vigour 'on-line' both implemented by multivariate data analysis. *Journal of the Institute of Brewing* 110: 3-17.

MUNCK, L., MØLLER, B., JACOBSEN, S. & SØNDERGAARD, S., 2004. Near infrared spectra indicate specific mutant endosperm genes and reveal a new mechanism for substituting starch with (1/3, 1/4)-b-glucan in barley. *J. Cereal Sci*. 40: 213-222.

MUNCK, L., NIELSEN J.P., MØLLER, B., JACOBSEN, S., SØNDERGAARD, I., ENGELSEN, S.B., NØRGAARD, L. & BRO, R., 2001. Exploring the phenotypic expression of a regulatory proteome-altering gene by spectroscopy and chemometrics. *Analytica Chimica Acta* 446: 171-186.

MUNCK, L., NORGAARD, L., ENGELSEN, S.B., BRO, B. & ANDERSSON, C.A., 1998. Chemometrics in food science-a demonstration of the feasibility of a highly exploratory, inductive evaluation strategy of fundamental scientific significance. *Chemometrics and Intelligent Laboratory Systems* 44: 31-60.

MÜNTZING, A., 1936. Über die Entstehungsweise 56-chromosomiger Weizen-Roggen-Bastarde. *Züchter* 8: 188-191.

MÜNTZING, A., 1939. Studies on the properties and ways of production of rye-wheat amhidiploids. *Hereditas* 25: 387-430.

MÜNTZING, A., 1957. Cytogenetic studies in rye-wheat (*Triticale*). *Cytologia* (suppl.) pp. 51-56.

MÜNTZING, A., 1963. Cytogenetic and breeding studies in Triticale. In: Proceedings of the Second International Wheat Genetics Symposium, Lund, Sweden. *Hereditas* (Suppl.) 2: 291-300.

MÜNTZING, A., 1979. Triticale. Results and Problems. *Fortschritte der Pflanzeniichtung. Zeitschrift fur Pflanzenzichtung, Beihefte* 10: 3-103.

MURAI, J., TAIRA, T. & OHTA, D., 1999. Isolation and characterization of three waxy genes encoding the granule-bound starch synthase in hexaploid wheat. *Gene* 234(1): 71-79.

MURAV'EVA, D.A., 1991. Farmakognoziya (in Russian). Moscow, Meditsina, 560 pp.

MURILLO, I., ROCA, R., BORTOLOTTI, C. & SEGUNDO, B., 2003. Engineering photoassimilate partitioning in tobacco plants improves growth and productivity and provides pathogen resistance. *Plant J.* 36: 330-341.

MURRAY, J.A.H. & CROCKETT, N., 1992. Antisense Techniques: An Overview. In: Murray, J.A.H. (Ed.), *Antisense RNA and DNA*. Wiley-Liss Inc., New York, pp. 1-49.

MYERS, A.M., MORELL, M.K., JAMES, M.G. & BALL, S.G., 2000. Recent progress toward understanding biosynthesis of the amylopectin crystal. *Plant Physiol*. 122: 989-997.

MYLLÄRINEN, P., SHULMAN, A.H., SALOVAARA, H. & POUTANEN, K., 1998. The effect of growth temperature on gelatinization properties of barley starch. *Acta Agric. Scand*. 48(1): 85-90.

NAES, T. & ISAAKSSON, T., 1992. How can outliers be detected in NIR spectroscopy? *NIR news* 3(5): 8-9.

NAGAMINE, T., YOSHIDA, H. & KOMAE, K., 1997. Varietal differences and chromosome locations of multiple isoforms of starch branching enzyme in wheat endosperm. *Phytochemistry* 46: 23-26.

NAKAMURA, Y., 2002. Towards a better understanding of the metabolic system for amylopectin biosynthesis in plants: rice endosperm as a model tissue. *Plant Cell Physiol*. 43: 718-725.

NAKAMURA, T., VRINTEN, P., HAYAKAWA, K. & IKEDA, J., 1998. Characterization of a granulo-bound starch synthase isoform found in the pericarp of wheat. *Plant Physiol*. 118: 125-132.

NAKAMURA, T., YAMAMORI, M., HIRANO, H. & HIDAKA, S., 1993a. Identification of three Wx proteins in wheat (*Triticum aestivum* L.). *Biochem. Genet.* 31: 75-86.

NAKAMURA, T., YAMAMORI, M., HIRANO, H. & HIDAKA, S., 1993b. The waxy (*Wx*) proteins of maize, rice and barley. *Phytochemistry* 33: 749-753.

NAKAMURA, T., YAMAMORI, M., HIRANO, H., HIDAKA, S. & NAGAMINE, T., 1995. Production of waxy (amylose-free) wheats. *Molecular and General Genetics* 248: 253-259.

NAKAMURA, Y., UMEMOTO, T., TAKAHATA, Y., KOMAE, K., AMANO, E. & SATOH, H., 1996. Changes in structure of starch and enzyme activities affected by sugary mutations in developing rice endosperm: possible role of starch debranching enzyme (R-enzyme) in amylopectin biosynthesis. *Physiol Plant* 97: 491-498.

NEBEL, B.R. & RUTTLE, M.L., 1938. The cytological and genetical significance of colchicine. *Hereditas* 29: 3-9.

NEBESNY, E., ROSICKA, J. & PIERZGALSKI, T., 1998. Enzymatic hydrolysis of wheat starch into glucose. *Starch/Stärke* 50(8): 337-341.

NEBESNY, E., ROSICKA, J. & TKACZYK, M., 2002. Effect of enzymatic hydrolysis of wheat starch on amylose-lipid complexes stability. *Starch/Stärke* 54: 603-608.

NEHRA, N.S., CHIBBAR, R.N., LEUNG, N., CASWELL, K., MALLARD, C., STEINHAUER, L., BÅGA, M. & KARTHA, K.K., 1994. Self-Fertile Transgenic Wheat Plants Regenerated from Isolated Scutellar Tissues Following Microprojectile Bombardment with Two Distinct Gene Constructs. *Plant J.* 5: 285-297.

NELSON, O.E., 1982. Genetic control of polysaccharide and storage protein synthesis in endosperms of barley, maize and sorghum. In: Y. Pomeranz (Ed.), *Advances in Cereal Science and Technology*. American Association of Cereal Chemists, St. Paul, pp. 41-71.

NELSON, O.E. & PAN, D., 1995. Starch synthesis in maize endosperm. *Annu. Rev. Plant Physiol. Plant Mol. Biol.* 46: 475-496.

NELSON, O.E. & RINES, H.W., 1962. The enzymatic deficiency in the waxy mutant of maize. *Biochem. Biophys. Res. Comm*. 9: 297-300.

NGONYAMO-MAJEE, D., 2005. Evaluation of corn endosperm properties and development of near infrared reflectance spectroscopy (NIRS) calibrations for screening germplasm on starch digestibility in large scale corn breeding programs. Ph.D. abstract, AAT 3186088, The University of Wisconsin, Madison, 236 p.

NIELSEN, J.P., PEDERSEN, D.K. & MUNCK, L., 2003. Development of nondestructive screening methods for single kernel characterization of wheat. *Cereal Chem*. 80(3): 274-280.

NIKOL'SKAYA, G.V., SOKOLOV, A.I., BATURINA, V.A. & GAPPAROV, M.M., 2001. Meditsinskie aspekty ispol'zovaniya enzim-rezistentnykh i modifitsirovannykh krakhmalov v pitanii cheloveka (in Russian). *Krakhmal i krakhmalosoderzhashchie istochniki – struktura, svojstva i novye tekhnologii:* Sbornik dokladov mezhdunarodnoj konferentsii. Moscow, p. 47.

NISHI, A., NAKAMURA, Y., TANAKA, N. & SATOH, H., 2001. Biochemical and genetic analysis of the effects of amylose-extender mutation in rice endosperm. *Plant Physiol*. 127: 459-472.

NODA, T., KIMURA, T., OTANI, M., IDETA, O., SHIMADA, T., SAITO, A. & SUDA, I., 2002. Physicochemical properties of amylose-free starch from transgenic sweet potato. *Carbohydr. Polym*. 49: 253-250.

NOLTE, B., 2006. Experiences on Bioethanol from Wheat. *2nd European Bioethanol Technology Meeting*. Retrieved November 4, 2007, from http://www.agfdt.de/loads/bi06/nolteabb.pdf

NOWOTNA, A., GAMBUŚ, H., KRATSCH, G., KRAWONTKA, J., GAMBUŚ, F., SABAT, R. & ZIOBRO, R., 2007. Effect of Nitrogen Fertilization on the Physico-chemical Properties of Starch Isolated from German Triticale Varieties. *Starch/Stärke* 59: 397-399.

NOZAKI, K., HAMADA, S., NAKAMORI, T., ITO, H., SAGISAKA, S., YOSHIDA, H., TAKEDA, Y., HONMA, M. & MATSUI, H., 2001. Major isoforms of starch branching enzymes in premature seeds of kidney bean (*Phaseolus vulgaris* L.). *Biosci. Biotechn. Biochem*. 65(5): 1141-1148.

O'SHEA, M.G., SAMUEL, M., KONIK, C.M. & MORELL, M., 1998. Flurophore assisted carbohydrate electrophoresis (FACE) of oligosaccharides: efficiency of labelling and high-resolution separation. *Carbohydr. Res.* 307: 1-12.

ODA, S. & SCHOFIELD, J.D., 1997. Characterization of friabilin polypeptides. *J. Cereal Sci*. 26: 29-36.

OELKE, E.A., OPLINGER, E.S. & BRINKMAN, M.A., 1989. Triticale. In: *Alternative Field Crops Manual*. Madison: University of Wisconsin Cooperative Extension, and St. Paul: University of Minnesota Extension Service. Retrieved November 7, 2007, from http://www.hort.purdue.edu/newcrop/afcm/triticale.html

OH, H.E., PINDER, D.N., HEMAR, Y., ANEMA, S.G. & WONG, M., 2008. Effect of high-pressure treatment on various starch-in-water suspensions. *Food Hydrocolloids* 22: 150-155.

OKECHUKWU, P. & RAO, M.A., 1995. Influence of granule size on viscosity of cornstarch suspension. *J. Texture Stud*. 26(5): 501-516.

OKUDA, M., ARAMAKI, I., KOSEKI, T., SATOH, H. & HASHIZUME, K., 2005. Structural characteristics, properties, and in vitro digestibility of rice. *Cereal Chem*. 82: 361-368.

OKUNO, K., 2005. Germplasm enhancement and breeding strategies for crop quality in Japan. *Plant Prod. Sci.* 8(3): 320-325.

ONDREY, G., 2005. Making bioethanol directly from starch. *Chemical Engineering* 112(4): 19(2).

OSBORNE, B.G., 2006. Applications of near infrared spectroscopy in quality screening of early-generation materials in cereal breeding programmes. *Journal of Near Infrared Spectroscopy* 14: 93-101.

OSBORNE, B.G., FAERN, T. & HINDLE, P.H., 1993. Practical NIR Spectroscopy with Applications in Food and Beverage Analysis. Second edition. Longman Scientific and Technical, Harlow, Essex, UK. 227 pp.

OSCARSSON, M., ANDERSSON, R., SALOMONSSON, A.-C. & AMAN, P., 1996. Chemical composition of barley samples focusing on dietary fiber components. *J. Cereal Sci*. 24: 161-169.

OVERTHROW, R. & CARVER, M.F., 2003. The value of triticale in the 2nd /3rd cereal position in crop sequences. *HGCA project report No. 306*. Home Grown Cereals Authority, Caledonia House, 223 Pentoville Rd, London.

PALIJ, A.F., 1989. Geneticheskie aspekty uluchsheniya kachestva zerna kukuruzy (in Russian). Kishinev, Shtiintsa, 175 pp.

PAN, D.D. & JANE, J., 2000. Internal structure of normal maize starch granules revealed by chemical surface gelatinization. *Biomacromolecules* 1: 126-132.

PAN, J., ZHU, Y. & CAO, W., 2007. Modelling plant carbon flow and grain starch accumulation in wheat. *Field Crops Research* 101: 276-284.

PANOZZO, J.F. & EAGLES, H.A., 1998. Cultivar and environmental effects on quality characters in wheat: I. Starch. *Aust. J. Agric Res*. 49: 757-766.

PARK, S.H., CHUNG, O.K. & SEIB, P.A., 2004. Size distribution and properties of wheat starch granules in relation to crumb grain score of pup-loaf bread. *Cereal Chem*. 81: 699-704.

PARK, S.H., CHUNG, O.K. & SEIB, P.A., 2005. Effects of varying weight ratios of large and small wheat starch granules on experimental straight-dough bread. *Cereal Chem*. 82: 166-172.

PARKER, M.L., 1985. The relationship between A-type and B-type starch granules in the developing endosperm of wheat. *J. Cereal. Sci*. 3: 271-278.

PARKER, R. & RING, S.G., 2001. Aspects of the physical chemistry of starch. *J. Cereal Sci.* 34: 1-17.

PASIKATAN, M.C. & DOWELL, F.E., 2004. High-Speed NIR Segregation of High- and Low-Protein Single Wheat Seeds. *Cereal Chem*. 81(1): 145-150.

PATZEK, T.W., 2004. Thermodynamics of the corn-ethanol biofuel cycle. *Critical Reviews in Plant Sciences* 23(6): 519-567. An updated web version is at http://petroleum.berkeley.edu/-papers/patzek/CRPS416-Patzek-Web.pdf

PATZEK, T.W., 2006a. A First-Law Thermodynamic Analysis of the Corn-Ethanol Cycle. *Natural Resources Research* 15(4): 255-270.

PATZEK, T.W., 2006b. A statistical analysis of the theoretical yield of ethanol from corn starch. *Natural Resources Research* 15(3): 205-212.

PATZEK, T.W., 2006c. Corn Ethanol: Laundering Fossil Fuels, Bilking Taxpayers, Damaging the Environment. *Energy Tribune*, April 2006, pp. 11-13.

PATZEK, T.W., ANTI, S.-M., CAMPOS, R., HA, K.W., LEE, J., LI, B., PADNICK, J. & YEE, S.-A., 2005. Ethanol from corn: Clean renewable fuel for the future, or drain on our resources and pockets? *Environment, Development & Sustainability* 7: 319-336.

PATZEK, T.W. & PIMENTEL, D., 2006. Thermodynamics of energy production from biomass. *Critical Reviews in Plant Sciences* 24(5-6): 327-364. Retrieved November 7, 2007, from http://petroleum.berkeley.edu/papers/patzek/CRPS-BiomassPaper.pdf

PAULSEN, M.R., PORDESIMO, L.O., SINGH, M., MBUVI, S.W. & YE, B., 2003. Maize Starch Yield Calibrations with Near Infrared Reflectance. *Biosystems Engineering* 85(4): 455-460.

PAULSEN, M.R. & SINGH, M., 2004. Calibration of a Near-infrared Transmission Grain Analyser for Extractable Starch in Maize. *Biosystems Engineering* 89(1): 79-83.

PAVLOVSKAYA, N.E., 2001. Razmer krakhmal'nykh granul i soderzhanie amilozy v protsesse razvitiya semyan gorokha (in Russian). *Krakhmal i krakhmalosoderzhashchie istochniki – struktura, svojstva i novye tekhnologii:* Sbornik mezhdunarodnoj konferentsii. Moscow, p. 166.

PEARSON, T.C., WICKLOW, D.T., MAGHIRANG, E.B., XIE, F. & DOWELL, F.E., 2001. Detecting aflatoxin in single corn kernels by transmittance and reflectance spectroscopy. *Transactions of the American Society of Agricultural Engineers* 44: 1247-1254.

PEARSON, T.C., WICKLOW, D.T. & PASIKATAN, M.C., 2004. Reduction of aflatoxin and fumonisin contamination in yellow corn by high-speed dual-wavelength sorting. *Cereal Chem.* 81: 490-498.

PEDERSEN, J.F., BEAN, S.R., FUNNELL, D.L. & GRAYBOSCH, R.A., 2004. Rapid iodine staining techniques for identifying the waxy phenotype in sorghum grain and waxy genotype in sorghum pollen. *Crop Sci*. 44: 764-767.

PEDERSEN, J.F., BEAN, S.R., GRAYBOSCH, R.A., PARK, S.H. & TILLEY, M. 2005. Characterization of waxy grain sorghum lines in relation to granule-bound starch synthase. *Euphytica* 144: 151-156.

PENA, R.J., TRETHOWAN, R., DFEIFFER, W.H. & VAN GINKEL, M., 2002. Quality (end use) improvement in wheat: compositional, genetic, and environmental factors. *J. Crop Prod*. 5: 1-37.

PENG, M., GAO, M., ABDEL-AAL, E.-S.M., HUCL, P. & CHIBBAR, R.N., 1999. Separation and characterization of A- and B-type starch granules in wheat endosperm. *Cereal Chem*. 76: 375-379.

PENG, M., GAO, M., BÅGA, M., HUCL, P. & CHIBBAR, R.N., 2000. Starch-Branching Enzymes Preferentially Associated with A-Type Starch Granules in Wheat Endosperm. *Plant Physiol.* 124: 265-272.

PERERA, C., LU, Z., SELL, J. & JANE, J., 2001. Comparison of physicochemical properties and structures of sugary-2 corn starch with normal and waxy cultivars. *Cereal Chem.* 78: 249-256.

PEREZ-VENDRELL, A.M., BRUFAU, J., MOLINA-CANO, J.L., FRANCESH, M. & GUASCH, J., 1996. Effects of cultivar and environment on b-(1-3, 1-4)-d-glucan content and acis extract viscosity of Spanish barleys. *J. Cereal Sci*. 23: 285-292.

PETERSON, D.G. & FULCHNER, R.G., 2001. Variation in Minnesota HRS wheats: starch granule size distribution. *Food Res. Intl.* 34: 357-363.

PIENAAR, R.D., 2006. History of breeding triticale at Stellenbosch University: the first 30 years. In: W.C. Botes, D. Boros, N. Darvey, P. Gustafson, R. Jessop, G.F. Marais, G. Oettler & D. Salmon (Eds.), *Proceedings of the 6th International Triticale Symposium*. 3-7 September 2006, Stellenbosch, South Africa, p. 14.

PIERNA, J.A.F. & DARDENNE, P., 2007. Chemometric contest at 'Chimiometrie 2005': A discrimination study. *Chemometrics and Intelligent Laboratory Systems* 86(2): 219-223.

PILLING, E. & SMITH, A.M., 2003. Growth ring formation in the starch granules of potato tubers. *Plant Physiol.* 132: 365-371.

PIMENTEL, D., 2003. Ethanol fuels: energy balance, economics, and environmental impacts are negative. *Natural Resources Research* 12(2): 127-134.

PIMENTEL, D., 2006. Soil erosion: A food and environment threat. *Environment, Development & Sustainability* 8: 119-137.

PIMENTEL, D. & PATZEK, T.W., 2005. Ethanol production using corn, switchgrass, and wood; biodiesel production using soybean and sunflower. *Natural Resource Research* 14(1): 67-76.

PISAREV, V. & VINOGRADOVA, N.M., 1944. Hybrids between wheat and Elymus. *Doklady Acad. Sci. USSR* 45(3).

PISAREV, V. & ZHILKINA, M.D., 1967. *Triticale* x 2n-42 (in Russian). *Genetika* 4: 3-12.

PLOCHL, M., HEIERMANN, M. & LINKE, B., 2003. Energy farming – potential and risk of biogas. In: *Jahresbericht*. Institut für Agrartechnik (Bornim) e.V., p. 73. Retrieved November 7, 2007, from http://www.atb-potsdam.de/hauptseite-deutsch/index2.htm

POLASKE, N.W., WOOD, A.L., CAMPBELL, M.R., NAGAN, M.C. & POLLAK, L.M., 2005. Amylose Determination of Native High-Amylose Corn Starches by Differential Scanning Calorimetry. *Starch/Stärke* 57(3-4): 118-123.

POLMAN, K., 1994. Review and analysis of renewable feedstocks for the production of commodity chemicals. Proc. 15th symposium on biotechnology for fuels and chemicals. Colorado Springs, USA, 10-14 May 1993. *Appl. Biochem. Biotechnol.* 45/46: 709-722.

POONI, H.S., KUMAR, I. & KHUSH, G.S., 1993a. Genetical control of amylose content in selected crosses of indica rice. *Heredity* 70: 269-280.

POTOP, V. & SOUKUP, J., 2009. Spatiotemporal characteristics of dryness and drought in the Republic of Moldova. *Theor. Appl. Climatol.* 96: 305-318.

POWER, R., 2003. Enzymatic conversion of starch to fermentable sugars. In: Jaques, K.A., Lyons, T.P., Kelsall, D.R. (Eds.), *The Alcohol Textbook*. Nottingham University Press, Nottingham UK.

PREISS, J., 1991. Biology and molecular biology of starch synthesis and its regulation. *Plant Molecular Cell Biol.* 7: 59-114.

PREISS, J. & LEVI, C., 1982. Starch biosynthesis and degradation. In: J. Preiss (Ed.), *The Biochemistry of Plants*, Vol. 3, *Carbohydrates: Structure and Function*. San Diego: Academic Press, pp. 371-423.

PREISS, J. & SIVAK, M.N., 1996. Starch synthesis in sinks and sources. In: Zamski E., Shaffer A.A., (Eds.), *Photoassimilate distribution and partitioning in plants and crops: source-sink relationships*. New York: Marcel Dekker, pp. 63-96.

PREISS, J. & SIVAK, M.N., 1998. Biochemistry, molecular biology and regulation of starch synthesis. In: L. Setlow (Ed.), *Genetic Engineering: Principles and Methods*, Vol. 20. Plenum Press, New York, pp. 177-223.

RAEKER, M.O., GAINES, C.S., FINNEY, P.L. & DONELSON, T., 1998. Granule size distribution and chemical composition of starches from 12 soft wheat cultivars. *Cereal Chem*. 75: 721-728.

RAGAUSKAS, A.J., WILLIAMS, C.K., DAVISON, B.H., BRITOVSEK, G., CAIRNEY, J., ECKERT, C.A., FREDERICK, W.J.JR., HALLETT, J.P., LEAK, D.J., LIOTTA, C.L., MIELENZ, J., MURPHY, R., TEMPLER, R. & TSCHAPLINSKI, T., 2006. The Path Forward for Biofuels and Biomaterials. *Science* 311: 484-489.

RAHMAN, S., ABRAHAMS, S., ABBOTT, D., MUKAI, Y., SAMUEL, M., MORELL, M. & APPELS, R., 1997. A Complex Arrangement of Genes at a Starch Branching Enzyme I Locus in the D-Genome Donor of Wheat. *Genome* 40: 465-474.

RAHMAN, S., BIRD, A., REGINA, A., LI, Z., RAL, J.P., MCMAUGH, S., TOPPING D. & MORELL, M., 2007. Resistant starch in cereals: Exploiting genetic engineering and genetic variation. *J. Cereal Sci.* 46(3): 251-260.

RAHMAN, S., KOSAR-HASHEMI, B., SAMUEL, M.S., HILL, A., ABBOTT, D.C., SKERRITT, J.H., PREISS, J., APPELS, R. & MORELL, M.K., 1995. The major proteins of wheat endosperm starch granules. *Australian Journal of Plant Physiology* 22: 793-803.

RAHMAN, S., LI, Z., BATEY, I., COCHRANE, M.P., APPELS, R. & MORELL, M., 2000. Genetic alteration of starch functionality in wheat. *J. Cereal Sci*. 31: 91-110.

RAHMAN, S., REGINA, A., LI, Z., MUKAI, Y., YAMAMOTO, M., KOSAR-HASHEMI, B., ABRAHAMS, S. & MORELL, M., 2001. Comparison of starch branching enzymes genes reveals evolutionary relationships among isoforms: characterisation of a gene for starch branching enzymes IIa from the wheat D genome donor *Aegilops tauschii*. *Plant Physiol.* 125: 1314-1324.

RAMESH, S., GOWDA, B.T.S. & GANGAPPA, E., 1998. Inheritance of qualitative characters in millet (*Panicum miliaceum* L.). *Crop Res*. 15(2/3): 218-220.

REDDY, A.N., RAO, K.V.K., SREENIVASULU, M.R., REDDY, V.C. & SAHEV, K.H., 1996. Genetic divergence in millet (*Panicum miliaceum* L.). *J. Res. APAU* 24(1/2): 92-95.

REGIERER, B., FERNIE, A.R., SPRINGER, F., PEREZ-MELIS, A., LEISSE, A., KOEHL, K., WILLMITZER, L., GEIGENBERGER, P. & KOSSMANN, J., 2002. Starch content and yield increase as a result of altering adenylate pools in transgenic plants. *Nat. Biotechnol.* 20: 1256-1260.

REGINA, A., BIRD, A., TOPPING, D., BOWDEN, S., FREEMAN, J., BARSBY, T., KOSAR-HASHEMI, B., LI, Z., RAHMAN, S. & MORELL, M., 2006. High-amylose wheat generated by RNA interference improves indices of large-bowel health in rats. *Proc. Natl. Acad. Sci. USA* 103: 3546-3551.

REGINA, A., KOSAR-HASHEMI, B., LI, Z., RAMPLING, L.R., CMIEL, M., GIANIBELLI, C., KONIK-ROSE, C., LARROQUE, O., RAHMAN, S. & MORELL, M.K., 2004. Multiple isoforms of starch branching enzyme-I in wheat: lack of the major SBE-I isoform does not alter starch phenotype. *Functional Plant Biology* 31: 591-601.

REICHARDT, M., 2007. Biofuels or food production: South Africa's dilemma. *ClimateChangeCorp.com*, 8 August 2007. Retrieved November 7, 2007, from http://www.climatechangecorp.com/content_print.asp?ContentID=4892

REIJNDERS, L. & HUIJBREGTS, M.A.J., 2007. Life cycle greenhouse gas emissions, fossil fuel demand and solar energy conversion efficiency in European bioethanol production for automotive purposes. *Journal of Cleaner Production* 15: 1806-1812.

RICHARDSON, P.H., JEFFCOAT, R. & SHI, Y.-C., 2000. High-amylose starches: from biosynthesis to their use as food ingredients. *MRS Bulletin* 25: 20-24.

RICHARDSON, T.H., XQ, T., FREY, G., CALLEN, W., CABELL, M., LAM, D., MACOMBER, J., SHORT, J.M., ROBERTSON, D.E. & MILLER, C., 2002. A novel, high performance enzyme for starch liquefaction – Discovery and optimization of a low pH, thermostable alpha-amylase. *J. Biol. Chem.* 277: 26501-26507.

RIFFKIN, H.L., BRINGHURST, T.A., MCDONALD, A.M.L. & HANDS, E., 1990. Quality requirements of wheat for distilling. *Aspects of Applied Biology* 25: 29-40.

RIKHTER, M., AUGUSTAT, Z. & SHIRBAUM, K., 1975. Izbrannye metody issledovaniya krakhmala (in Russian). Moscow, Pishchevaya promyshlennost', 183 pp.

RINDLAV-WESTLING, A., STADING, M. & GATENHOLM, P., 2002. Crystallinity and morphology in films of starch, amylose and amylopectin blends. *Biomacromolecules* 3: 84-91.

RITTE, G., LLOYD, J.R., ECKERMANN, N., ROTTMANN, A., KOSSMANN, J. & STEUP, M., 2002. The starch-related R1 protein is an alpha-glucan, water dikinase. *Proc. Natl. Acad. Sci. USA* 99: 7166-7171.

RODRÍGUEZ-LÓPEZ, M., BAROJA-FERNÁNDEZ, E., ZANDUETA-CRIADO, A. & POZUETA-ROMERO, J., 2000. Adenosine diphosphate glucose pyrophosphatase: a plastidial phosphodiesterase which prevents starch biosynthesis. *Proc. Natl. Acad. Sci. USA* 97: 8705-8710.

RODRIGUEZ-QUIJANO, M., VAZQUEZ, J.F. & CARRILLO, J.M., 2004. Waxy proteins and amylose content in diploid *Triticeae* species with genomes A, S and D. *Plant Breeding* 123: 294-296.

ROHDE, W., BECKER, D. & SALAMINI, F., 1988. Structural analysis of the waxy locus from *Hordeum vulgare. Nucleic acids research* 16(14): 7185-7186.

ROSENBERGER, A., 2005. Identification of top-performing cereal cultivars for grain-to-ethanol operations. *Zuckerindustrie* 130(9): 697-701.

ROSENBERGER, A., KAUL, H.-P., SENN, T. & AUFHAMMER, W., 2000. Optimierung der Produktion von Wintergetreide zur Bioethanolherstellung durch unterschiedlich intensive Anbauverfahren. (Improving the production of winter cereals for bioethanol by cultivation with varied intensity). *J. Agron. Crop Sci.* 185(1): 55-65.

ROSENBERGER, A., KAUL, H.P., SENN, T. & AUFHAMMER, W., 2002. Costs of bioethanol production from winter cereals: the effect of growing conditions and crop production intensity levels. *Indust. Crops Prod*. 15: 91-102.

ROSS, A.S., QUAIL, K.J. & CROSBIE, G.B., 1996. An insight into structural features leading to desirable alkaline noodle texture. In: *Proc. 46th Australian Cereal Chemistry Conference*. Sydney, Australia, pp. 115-119.

ROUX, H.S., MARAIS, G.F., SNYMAN, J.E. & BOTES, W.C., 2006. The South African Triticale breeding programme: a report on recent developments. In: W.C. Botes, D. Boros, N. Darvey, P. Gustafson, R. Jessop, G.F. Marais, G: Oettler & D. Salmon (Eds.), *Proceedings of the 6th International Triticale Symposium*. 3-7 September 2006, Stellenbosch, South Africa, p. 32.

ROWLAND-BAMFORD, A.J., ALLEN, L.H.JR., BAKER, J.T. & BOOTE, K.J., 1990. Carbon dioxide effects on carbohydrate status and partitioning in rice. *Plant Cell Environ*. 14: 1601-1608.

RUDI, H., DOAN, D.N. & OLSEN, O.A., 1997. A (His)6-tagged recombinant barley (*Hordeum vulgare* L.) endosperm ADP- glucose pyrophosphorylase expressed in the baculovirus-insect cell system is insensitive to allosteric regulation by 3-phosphoglycerate and inorganic phosphate. *FEBS Lett*. 419: 124-130.

RUDI, H., UHLEN, A.K., HARSTAD, O.M. & MUNCK, L., 2006. Genetic variability in cereal carbohydrate compositions and potentials for improving nutritional value. *Animal Feed Science and Technology* 130: 55-65.

RYABCHOUN, V.K. & SHATOKHIN, V.I., 2005. Selektsiya yarogo triticale v Ukraini (in Ukrainian). In: *Naukove zabezpechennya vyrobnytstva zerna triticale i produktiv jogo pererobky:* Tezisy mizhnarodnoi naukovo-praktichnoi konferentsii, 6-8 lypnya 2005 r. Kharkiv, Instytut roslynnytstva im. V.Ya. Yur'eva UAAN, pp. 27-28.

RYABCHOUN, V.K., SHATOKHIN, V.I., LISNICHIJ, V.A. & KAPUSTINA, T.B., 2007. Yare tritikale dlya stabil'nogo virobnitstva zerna (in Ukrainian). Kharkiv, Institut roslinnitstva im. V.Ya. Yur'eva, 16 pp.

RYABCHOUN, V.K., SHATOKHIN, V.I., LISNICHIJ, V.A., KAPUSTINA, T.B. & KURTASOV, B.V., 2006. Katalog sortiv yarogo triticale selektsii Instytutu roslynnytstva im. V.Ya. Yur'eva (in Ukrainian). Kharkiv, Instytut roslynnytstva im. V.Ya. Yur'eva, 22 pp.

SAFFORD, R., JOBLING, S.A., SIDEBOTTOM, C.M., WESTCOTT, R.J., COOKE, D., TOBER, K.J., STRONGITHARM, B.H., RUSSELL, A. & GIDLEY, M.J., 1998. Consequences of antisense RNA inhibition of starch branching enzyme activity on properties of potato starch. *Carbohyd. Polym*. 35: 155-168.

SAHLSTRÖM, S., BÆVRE, A.B. & GRAYBOSCH, R., 2006. Impact of Waxy, Partial Waxy, and Wildtype Wheat Starch Fraction Properties on Hearth Bread Characteristics. *Cereal Chem.* 83(6): 647-654.

SAHLSTRÖM, S., BRÆTHEN, E., LEA, P. & AUTIO, K., 1998. Influence of starch granule size distribution on bread characteristics. *J. Cereal Sci*. 28: 157-164.

SAITO, K., ITO, T., KURIBAYASHI, T., MOCHIDA, T., NAKAKUKI, T., SHIBATA, M. & SUGAWARA, M., 2001. Effect of raw and heat-moisture treated high-amylose corn starch on fermentation by the rat cecal bacteria. *Starch/Stärke* 53: 424-430.

SAJILATA, M.G., SINGHAL, R.S. & KULKARNI, P.R., 2006. Resistant Starch – A Review. *Comprehensive Reviews in Food Science and Food Safety* 5: 1-17.

SAKAMOTO, T. & MATSUOKA, M., 2004. Generating high-yielding varieties by genetic manipulation of plant architecture. *Curr Opin. Biotechnol*. 15: 144-147.

SAKULSINGHAROJ, C., CHOI, S.-B., HWANG, S.-K., EDWARDS, G.E., BORK, J., MEYER, C.R., PREISS, J. & OKITA, T.W., 2004. Engineering starch biosynthesis for increasing seed weight: the role of the cytoplasmic ADP-glucose pyrophosphorylase. *Plant Sci.* 167: 1323-1333.

SALOMONSSON, A.C. & SUNDBERG, B., 1994. Amylose content and chain profile of amylopectin from normal, high amylose and waxy barleys. *Starch/Stärke* 46: 325-328.

SANCHEZ, P.C., JULIANO, B.O., LAUDE, V.T. & PEREZ, C.M., 1988. Nonwaxy Rice for Tapuy (Rice Wine) Production. *Cereal Chem*. 65(3): 240-243.

SANCHEZ-MONGE, E. & TJIO, J.H., 1954. Note on a 42-chromosome Triticale. *Proc. 9th Int. Cong. Genet. Carylogia* Suppl. 6: 748.

SANDSTEDT, R.M., HITES, B.D. & SCHROEDER, H., 1968. Genetic variations in maize: Effect on properties of starches. *Cereal. Sci. Today* 13: 82-85.

SANDSTEDT, R.M., STRAHAN, D., UEDA, S. & ABBOT, R.C., 1962. The digestibility of high-amylose corn starches compared to that of other starches. The apparent effect of ae gene on susceptibility to amylase action. *Cereal Chem*. 39: 123-131.

SANO, Y., KATSUMATA, M. & OKUNO, K., 1986. Genetic studies of speciation in cultivated rice. 5. Inter- and intra-specific differentiation in the waxy gene expression of rice. *Euphytica* 35: 1-9.

SANO, Y., MAEKAWA, M. & KIKUCHI, H., 1985. Temperature effects on the *Wx* protein level and amylose content in the endosperm of rice. *J. Hered*. 6: 221-222.

SARATH, G., MITCHELL, R.B., SATTLER, S.E., FUNNELL, D., PEDERSEN, J.F., GRAYBOSCH, R.A. & VOGEL, K.P., 2008. Opportunities and roadblocks in utilizing forages and small grains for liquid fuels. *J. Ind. Microbiol. Biotechnol*. 35: 343-354.

SASAKI, T., YASUI, T., MATSUKI, J. & SATAKE, T., 2002. Comparison of wheat starch gels with different amylose content. *Cereal Chem*. 79: 861-866.

SATHISH, P., SUN, C., LONNEBORG, A. & JANSSON, C., 1995. Modified starch metabolism in mutant and transgenic plants. *Prog. Bot.* 56: 301-318.

SATOH, H., NISHI, A., YAMASHITA, K., TAKEMOTO, Y., TANAKA, Y., HOSAKA, Y., SAKURAI, A., FUJITA, N. & NAKAMURA, Y., 2003. Starch branching enzyme-I-deficient mutation specifically affects the structure and properties of starch in rice endosperm. *Plant Physiol*. 133: 1111-1121.

SAULNIER, L., PENEAU, N. & THIBAULT, J.F., 1995. Variability in grain extract viscosity and watersoluble arabinoxylan content in wheat. *J. Cereal Sci.* 22: 259-264.

SAWYER, D., 2008. Climate change, biofuels and eco-social impacts in the Brazilian Amazon and Cerrado. *Phil. Trans. R. Soc. B.* 363: 1747-1752.

SCHÄFER, V., AUFHAMMER, W., KÜBLER, E., PIEPER, H.-J. & SENN, T., 1997. Energiebilanzen zur Produktion und Verarbeitung des Kornguts von Wintertriticale und Winterweizen zu Bioethanol. *Pflanzenbauwissenschaften* 1(3): 133-141.

SCHULMAN, A.H., 1999. Chemistry, biosynthesis, and engineering of starches and other carbohydrates. In: Paredes-Lopez, O. (Ed.), *Molecular biotechnology for plant food production*. Technomic Publishing Co., Inc., Lancaster, A, USA, pp. 493-523.

SCHULMAN, A.H. & AHOKAS, H., 1990. A novel shrunken endosperm mutant of barley. *Physiol. Plant* 78: 583-589.

SCHWALL, G.P., SAFFORD, R., WESTCOTT, R.J., JEFFCOAT, R., TAYAL, A., SHI, Y.C., GIDLEY, M.J. & JOBLING, S.A., 2000. Production of very-high-amylose potato starch by inhibition of SBE A and B. *Nat. Biotechnol*. 18(5): 551-554.

SCHWENDER, J., GOFFMAN, F., OHLROGGE, J. & SHACHAR-HILL, Y., 2004. Rubisco without the Calvin cycle improves the carbon efficiency of developing green seeds. *Nature* 432: 779-782.

SCHWENDER, J., OHLROGGE, J. & SHACHAR-HILL, Y., 2004. Understanding flux in plant metabolic networks. *Curr. Opin. Plant Biol*. 7: 309-317.

SEARCHINGER, T., HEIMLICH, R., HOUGHTON, R.A., DONG, F., ELOBEID, A., FABIOSA, J., TOKGOZ, S., HAYES, D. & YU, T.-H., 2008. Use of U.S. Croplands for Biofuels Increases Greenhouse Gases Through Emissions from Land-Use Change. *Science* 319: 1238-1240.

SEHNKE, P.C., CHUNG, H.J., WU, K. & FERL, R.J., 2001. Regulation of starch accumulation by granule-associated plant 14-3-3 proteins. *Proc. Natl. Acad. Sci. USA* 98: 765-770.

SEIB, P.A., 1994. Wheat starch: isolation, structure and properties. *Oyo Toshitsu Kagaku* 41: 49-69.

SEKULIC, S.S., WAKEMAN, J., DOHERTY, P. & HAILEY, P.A., 1998. Automated system for the on-line monitoring of powder blending processes using near-infrared spectroscopy. Part II. Qualitative approaches to blend evaluation. *Journal of Pharmaceutical and Biomedical Analysis* 17: 1285-1309.

SELYANINOV, G.T., 1958. The nature and dynamics of the droughts. In: Rudenko A.I. (ed.), *Droughts in the USSR. Their Nature, Recurrences, and Impact on Crops Yields* (in Russian). Gidrometeoizdat, Leningrad, pp. 5-29.

SEMEIJN, C. & BUWALDA, P., 2006. Eliane, a New Waxy Potato Starch. *The 57th Starch Convention*. Retrieved November 4, 2007, from http://www.agfdt.de/loads/st06/semeijn.pdf

SENE, M., CAUSSE, M., DAMERVAL, C., THEVENOT, C. & PRIOUL, J.-L., 2000. Quantitative trait loci affecting amylose, amylopectin and starch content in maize recombinant inbred lines. *Plant Physiol. Biochem*. 38(6): 459-472.

SENEVIRATNE, H.D. & BILIADERIS, C.G., 1991. Action of a-amylases on amylose-lipid complex superstructures. *J. Cereal Sci*. 13: 129-141.

SENN, T., 2000. Aspekte der technologischen Nutzung von Triticale. *Vorträge für Pflanzenzüchtung* 49: 179-186.

SENN, T. & PIEPER, H.J., 2000. Classical methods. In: Roehr, M. (Ed.), *The biotechnology of ethanol: classical and future applications*. Wiley-VCH, Weinheim, Germany, p. 1-84.

SENN, T. & PIEPER, H.J., 2001. Classical methods. In: Roehr, M. (Ed.), *The Biotechnology of Ethanol: Classical and Future Applications*. WILEY-VCH Verlag GmbH, Weinheim, New York, Chichester, Brisbane, Singapore, Toronto, p. 74-75.

SENN, T., THOMAS, L. & PIEPER, H.J., 1993. Zur Bedeutung autoamylolytiscer Eigenschaften von Triticale-Sorten fűr die Garungsalkoholgewinnung, *Handb. Brennerei Alkoholwirtsch*. 40: 383-401.

SERBIN, A.G., SERAYA, L.M., TKACHENKO, N.M. & SLOBODYANYUK, T.A., 2003. Meditsinskaya botanika (in Russian). Kharkov, NFAU Zolotye stranitsy, 364 pp.

SHANNON, J.C., CREECH, R.G. & LOERCH, J.D., 1970. Starch synthesis studies in Zea mays: II. Molecular distribution of radioactivity in starch. *Plant Physiol*. 45: 163-168.

SHANNON, J.C. & GARWOOD, D.L., 1984. Genetics and physiology of starch development. In: Whistler (Ed.), *Starch: Chemistry and Technology*. 2nd ed., pp. 25-79.

SHANNON, J.C., PIEN, F.M. & LIU, K.C., 1996. Nucleotides and nucleotide sugars in developing maize (*Zea mays* L.) endosperms: Synthesis of ADP-glucose in brittle-1. *Plant Physiol*. 110: 835-843.

SHARIFLOU, M.R. & SHARP, P.J., 1999. A polymorphic microsatellite in the 3' end of 'waxy' genes of wheat, *Triticum aestivum*. *Plant Breeding* 118: 275-277.

SHARMA, V., RAUSCH, K.D., TUMBLESON, M.E. & SINGH, V., 2007. Comparison Between Granular Starch Hydrolyzing Enzyme and Conventional Enzymes for Ethanol Production from Maize Starch with Different Amylose: Amylopectin Ratios. *Starch/Stärke* 59: 549-556.

SHARMA, R., SISSON, M.J., RATHJEN, A.J. & JENNER, C.F., 2002. The null 4A allele at the waxy locus in durum wheat affects pasta cooking quality. *J. Cereal Sci*. 35: 387-297.

SHENK, J.S. & WESTERHAUS, M.O., 1996. Calibration the ISI way. In: Davies, A.M.C., Williams, P.C. (Eds.), *Near Infrared Spectroscopy: The Future Waves*. NIR Publications, Chichester, UK. pp. 198–202.

SHETTY, J.K., LANTERO, O.J. & DUNN-COLEMAN, N., 2005. Technological advances in ethanol production. *Int. Sugar. J*. 107: 605-606, 608.

SHI, Y.-C., SEIB, P. & BERNARDIN, J.E., 1994. Effects of temperature during grain filling on starches from six wheat cultivars. *Cereal Chem*. 71(3): 369-383.

SHIBANUMA, K., TAKEDA, Y., HIZUKURI, S. & SHIBATA, S., 1994. Molecular structures of some wheat starches. *Carbohyd. Polym.* 25: 111-116.

SHIGECHI, H., KOH, J., FUJITA, Y., MATSUMOTO, T., BITO, Y., UEDA, M., SATOH, E., FUKUDA, H. & KONDO, A., 2004. Direct production of ethanol from raw corn starch via fermentation by use of a novel surface-engineered yeast strain codisplaying glucoamylase and a-amylase. *Appl. Environ. Microbiol*. 70(8): 5037-5040.

SHIMADA, H., TADA, Y., KAWASAKI, T. & FUJIMURA, T., 1993. Antisense Regulation of the Rice waxy Gene Expression Using a PCR-Amplified Fragment of the Rice Genome Reduces the Amylose Content in Grain Starch. *Theor. Appl. Genet*. 86: 665-672.

SHINDE, S.V., NELSON, J.E. & HUBER, K.C., 2003. Soft wheat starch pasting behavior in relation to A- and B-type granule content and composition. *Cereal Chem*. 80: 91-98.

SHITS, L.A., 2001. Krakhmal'nye reagenty v tekhnologii burovykh rastvorov: traditsii i perspektivy (in Russian). *Krakhmal i krakhmalosoderzhashchie istochniki – struktura, svojstva i novye tekhnologii:* Sbornik dokladov mezhdunarodnoj konferentsii. Moscow, p. 55.

SHLENS, J., 2009. A Tutorial on Principal Component Analysis. Version 3.01. April 22, 2009. Center for Neural Science, New York University and Systems Neurobiology Laboratory, Salk Institute for Biological Studies, pp. 1-12. Retrieved May 3, 2009, from http://www.snl.salk.edu/~shlens/pub/notes/pca.pdf

SHMARAEV, G.E., 1975. Kukuruza (in Russian). Moscow, Kolos, 303 pp.

SHONDELMAIER, J., JACOBI, J., FISCHBECK, G. & JAHOOR, A., 1992. Genetic studies on the mode of inheritance and localization of the *amol* (high amylose) gene in barley. *Plant Breeding* 109(2): 274-290.

SHURE, M., WESSLER, S. & FEDOROFF, N., 1983. Molecular identification and isolation of the waxy locus in maize. *Cell* 35: 225-233.

SIDEBOTTOM, C., KIRKLAND, M., STRONGITHARM, B. & JEFFCOAT, R., 1998. Characterisation of the difference of starch branching enzyme activities in normal and low-amylopectin maize during kernel development. *J. Cereal Sci*. 27: 279-287.

SIEVERT, D. & POMERANZ, Y., 1989. Enzyme-resistant starch. I. Characterization and evaluation by enzymatic, thermoanalytical, and microscopic methods. *Cereal Chem*. 66(4): 342-347.

SIKKA, V.K., CHOI, S.-B., KAVAKLI, I.H.I.H., SAKULSINGHAROJ, C., GUPTA, S., ITO, H. & OKITA, T.W., 2001. Subcellular compartmentation and allosteric regulation of the rice endosperm ADPglucose pyrophosphorylase. *Plant Sci*. 161: 461-468.

SIMMONDS, N.W., 1995. The relation between yield and protein in cereal grain. *J. Sci. Food Agric.* 67: 309-315.

SINGH, M., PAULSEN, M.R., TIAN, L. & YAO, H., 2002. Site-specific study of corn protein, oil, and extractable starch variability using NIT spectroscopy. *ASAE Paper No. 02-1111*. Retrieved November 4, 2007, from http://asae.frymulti.com/abstract.asp?aid=9149&t=2

SINGLETARY, G.W., BANISADR, R. & KEELING, P.L., 1994. Heat stress during grain filling in maize: Effects on carbohydrate storage and metabolism. *Aust. J. Plant Physiol*. 21: 829-841.

SISSONS, M., OSBORNE, B. & SISSONS, S., 2006. Application of near infrared reflectance spectroscopy to a durum wheat breeding programme. *J. Near Infrared Spectrosc.* 14: 17-25.

SLADE, A.J., FUERSTENBERG, S.I., LOEFFLER, D., STEINE, M.N. & FACCIOTTI, D., 2005. A reverse genetic, nontransgenic approach to wheat crop improvement by TILLING. *Nature Biotechnology* 23: 75-81.

SLATTERY, C.J., KAVAKLI, I.H. & OKITA, T.W., 2000. Engineering starch for increased quantity and quality. *Trends Plant Sci.* 5(7): 291-298.

SMAIL, V.W., FRITZ, A.K. & WETZEL, D.L., 2006. Chemical imaging of intact seeds with NIR focal plane array assists plant breeding. *Vibrational Spectroscopy* 42(2): 215-221.

SMIDANSKY, E.D., CLANCY, M., MEYER, F.D., LANNING, S.P., BLAKE, N.K., TALBERT, L.E. & GIROUX, M.J., 2002. Enhanced ADP-glucose pyrophosphorylase activity in wheat endosperm increases seed yield. *Proc. Natl. Acad. Sci. USA* 99: 1724-1729.

SMIDANSKY, E.D., MARTIN, J.M., HANNAH, L.C., FISCHER, A.M. & GIROUX, M.J., 2003. Seed yield and plant biomass increases in rice are conferred by deregulation of endosperm ADP-glucose pyrophosphorylase. *Planta* 216: 656-664.

SMITH, A.M., 1988. Major differences in isoforms of starch-branching enzyme between developing embryos of round- and wrinkled-seeded peas. *Planta* 175: 170-179.

SMITH, A.M., 1999. Making starch. *Curr. Opin. Plant Biol*. 2: 223-229.

SMITH, A.M., 2001. The biosynthesis of starch granules. *Biomacromolecules* 2: 335-341.

SMITH, A.M., BETTEY, M. & BEDFORD, I.D., 1989. Evidence that the *rb* locus alters the starch content of developing pea embryos through an effect on ADP glucose pyrophosphorylase. *Plant Physiol.* 89: 1279-1284.

SMITH, A.M., DENYER, K. & MARTIN, C., 1995. What controls the amount and structure of starch in storage organs? *Plant Physiol.* 107: 673-677.

SMITH, A.M., DENYER, K. & MARTIN, C., 1997. The synthesis of the starch granule. *Ann. Rev. Plant Physiol. Plant Mol. Biol.* 48: 67-87.

SMITH, T.C., KINDRED, D.R., BROSNAN, J.M., WEIGHTMAN, R.M., SHEPHERD, M. & SYLVESTER-BRADLEY, R., 2006. Wheat as a feedstock for alcohol production. *HGCA Research review No. 61*. Home Grown Cereals Authority, Caledonia House, 223 Pentoville Rd, London. Retrieved November 4, 2007, from http://www.lowcvp.org.uk/assets/reports/HGCA%20RR61%20Wheat%20for%20alcohol.pdf

SMITH, A.M. & MARTIN, C., 1993. Starch biosynthesis and the potential for its manipulation. In: D. Grierson (Ed.), *Biosynthesis and Manipulation of Plant Products. Plant Biotechnology*. Glasgow: Blackie and Son Publ., pp. 1-54.

SOHN, M., BARTON, F.E., MCCLUNG, A.M. & CHAMPAGNE, E.T., 2004. Near-infrared spectroscopy for determination of protein and amylose in rice flour through use of derivatives. *Cereal Chemistry* 81: 341-344.

SONG, H., DELWICHE, S.R. & CHEN, Y.R., 1995. Neural network single kernel near-infrared transmittance classification of wheat using spectra. *Optical Eng.* 34: 2927-2934.

SONG, Y. & JANE, J., 2000. Characterization of barley starches of waxy, normal, and high amylose varieties. *Carb. Polym.* 41: 365-377.

SONNEWALD, U., HAJIREZAEI, M., KOSSMANN, J., HEYER, A., TRETHEWEY, R. & WILLMITZER, L., 1997. Increased potato tuber size resulting from apoplastic expression of a yeast invertase. *Nat. Biotechnol*. 15: 794-797.

SORENG, R.J., 2003. *Triticosecale*. In: Catalogue of New World Grasses (*Poaceae*): IV. Subfamily *Pooideae*. *Contr. U.S. Natl. Herb*. 48: 676.

SORENSEN, H.R., PEDERSEN, S. & MEYER, A.S., 2006. Optimization of reaction conditions for enzymatic viscosity reduction and hydrolysis of wheat arabinoxylan in an industrial ethanol fermentation residue. *Biotechnology Progress* 22: 505-513.

SOSULSKI, K. & TARASOFF, L., 1997. Evaluation of cereal grains as a feed stock for ethanol production. In: Stumborg, M.A. (Ed.), *Proc. 1997 Ethanol research and development workshop*. Pub. NRC/AAFC, pp. 58-61.

SOZINOV, A.A. & ZHEMELA, G.P., 1983. Uluchshenie kachestva zerna ozimoj pshenitsy i kukuruzy (in Russian). Moscow, Kolos, 270 pp.

SPENCE, K.E. & JANE, J., 1999. Chemical and physical properties of ginkgo (*Ginkgo biloba*) starch. *Carbohydr. Polym*. 40: 261-269.

STACE, C.A., 1987. Triticale: a case of nomenclatural mistreatment. *Taxon* 36(2): 445-452.

STAERK, E. & GRUNEWALD, H., 2006. Utilisation of Near Infrared Spectroscopy in the Bioethanol Industry. *2nd European Bioethanol Technology Meeting*. Retrieved November 4, 2007, from http://www.agfdt.de/loads/bi06/staerk.pdf

STAHL, Y., COATES, S., BRYCE, J.H. & MORRIS, P.C., 2004. Antisense downregulation of the barley limit dextrinase inhibitor modulates starch granule size distribution, starch composition and amylopectin structure. *Plant J.* 39: 599-611.

STALEY, B.C. & BRADLEY, R., 2008. Plants at the Pump: Reviewing Biofuels Impacts and Policy Recommendations. World Resources Institute, *Climate and Energy (Policy Series).* July 2008, pp. 1-8. Retrieved November 16, 2008, from http://pdf.wri.org/plants_at_the_pump_brief.pdf

STARK, D.M., TIMMERMAN, K.P., BARRY, G.F., PREISS, J. & KISHORE, G.M., 1992. Regulation of the amount of starch in plant tissues by ADP glucose pyrophosphorylase. *Science* 258: 287-292.

STEFFENS, K.-J., 2006. Application of Starches, Modified Starches and Starch Derivatives in Pharmaceutical Products. *The 57th Starch Convention*. Retrieved November 4, 2007, from http://www.agfdt.de/loads/st06/steffens.pdf

STEVNEBØ, A., SAHLSTRØM, S. & SVIHUS, B.S., 2006. Starch structure and degree of starch hydrolysis of small and large starch granules from barley varieties with varying amylose content. *Anim. Feed Sci. Technol*. 130: 23-38.

STINARD, P.S., ROBERTSON, D.S. & SCHNABLE, P.S., 1993. Genetic isolation, cloning and analysis of a Mutator-induced, dominant antimorph of maize amylose extender 1 locus. *Plant Cell*. 5: 1555-1566.

STITT, M., 1996. Plasmodesmata play an essential role in sucrose export from leaves: a step toward an integration of metabolic biochemistry and cell biology. *Plant Cell*. 8: 565-571.

STODDARD, F.L., 1999. Survey of starch particle-size distribution in wheat and related species. *Cereal Chem*. 76: 145-149.

STUTE, R., KLINGLER, R.W., BOGUSLAWSKI, S., ESHTIAGHI, M.N. & KNORR, D., 1996. Effects of high pressures treatment on starches. *Starch/Stärke* 48: 399-408.

SUBBARAO, G.V., TOMOHIRO, B., MASAHIRO, K., OSAMU, I., SAMEJIMA, H., WANG, H.Y., PEARSE, S.J., GOPALAKRISHNAN, S., NAKAHARA, K., ZAKIR HOSSAIN, A.K.M., TSUJIMOTO, H. & BERRY, W.L., 2007. Can biological nitrification inhibition (BNI) genes from perennial *Leymus racemosus* (*Triticeae*) combat nitrification in wheat farming? *Plant & Soil* 299: 55-64.

SUH, D.S., AO, Z. & IANE, I.-L., 2001. Effects of starch structures on its application. *Krahmal i krahmalosoderzhashchie istochniki – struktura, svojstva i novye tehnologii.* Sbornik dokladov mezhdunarodnoj konferencii (in Russian). Moscow, p. 37.

SUN, J., OKITA, T.W. & EDWARDS, G.E., 1999. Modification of Carbon Partitioning, Photosynthetic Capacity, and O_2 Sensitivity in Arabidopsis Plants with Low ADP-Glucose Pyrophosphorylase Activity. *Plant Physiol*. 119: 267-276.

SUN, C., SATHISH, P., AHLANDSBERG, S. & JANSSON, C., 1997. Identification of four starch-branching enzymes in barley endosperm: partial purification of forms I, IIa and IIb. *New Phytol*. 137: 215-222.

SUN, C., SATHISH, P., AHLANDSBERG, S. & JANSSON, C., 1999. Analyses of isoamylase gene activity in wild-type barley indicate its involvement in starch synthesis. *Plant Mol. Biol*. 40: 431-443.

SVIHUS, B., UHLEN, A.K. & HARSTAD, O.M., 2005. Effect of starch granule structure, associated components and processing on nutritive value of cereal starch: A review. *Animal Feed Science and Technology* 122: 303-320.

SWANSTON, J.S. & NEWTON, A.C., 2005. Mixtures of UK wheat as an efficient and environmentally friendly source for bioethanol. *Journal of Industrial Ecology* 9: 109-126.

SWANSTON, J., NEWTON, A., BROSNAN, J., FOTHERINGHAM, A. & GLASGOW, E., 2005. Determining the spirit yield of wheat varieties and variety mixtures. *J. Cereal Sci.* 42: 127-134.

SWANSTON, J.S., NEWTON, A.C. & SMITH, P.L., 2006. Grain quality characters in complex mixtures of soft wheat and acknowledgement by farmers and distillers. In: Østergård, H., Fontaine, L. (Eds.), *Proceedings of the COST SUSVAR workshop on Cereal crop diversity: Implications for production and products*. Domaine de La Besse (Camon, Ariège), France, 13-14 June 2006. ITAB, Paris, France, 153 pages, p. 29-32.

SWANSTON, J.S. & SMITH, P.L., 2008. A turbidity test for a genetic component of spirit yield in wheat. In: Bryce, J., Piggott, J. and Stewart, G.G. (Eds.), *Distilled Spirits: Production Technology and Innovation*. Nottingham University.

SWANSTON, J.S., SMITH, P.L., GILLESPIE, T.L., BROSNAN, J.M., BRINGHURST, T.A. & AGU, R.C., 2007. Associations between grain characteristics and alcohol yield among soft wheat varieties. *J. Sci. Food Agric*. 87: 676-683.

SYLVESTER-BRADLEY, R., 2007. GREEN Grain. *Biofuels International* 1: 37-39.

SYLVESTER-BRADLEY, R. & KINDRED, D.R., 2008. Developing and growing wheat for the biofuels market. In: *Arable cropping in a changing climate*. HGCA conference, 23 and 24 January 2008, pp. 110-125.

TAKATA, K., NISHIO, Z., IRIKI, N., TABIKI, T., FUNATSUKI, W. & YAMAUCHI, H., 2005. Comparison of quality characteristics of waxy wheat using a near isogenic line. *Breed. Sci.* 55: 87-92.

TAKATA, K., YANAKA, M., FUJITA, Y. & ISHIKAWA, N., 2007. Evaluation of the Grain and Flour Quality in Near-Isogenic Wheat Lines with Waxy and Double-null *Wx* proteins. *Breed. Sci.* 57: 79-83.

TAKAHA, T., YANASE, M., OKADA, S. & SMITH, S.M., 1993. Disproportionating enzyme (4-α-glucanotransferase; EC 2.4.1.25) of potato. *J. Biol. Chem*. 268: 1392-1396.

TAKEDA, Y., GUAN, H.P. & PREISS, J., 1993. Branching of amylose by branching isoenzymes of maize endosperm. *Cabohydr. Res*. 240: 253-263.

TAKEDA, Y. & HIZUKURI, S., 1987. Structures of branched molecules of amyloses of various origins, and molar fractions of branched and unbranched molecules. *Carbohydr. Res.* 165: 139-145.

TAKEDA, Y., HIZUKURI, S. & JULIANO, B.O., 1987. Structures of rice amylopectins with low and high affinities for iodine. *Carbohydr. Res*. 168: 79-88.

TAKEDA, Y., SHIRASAKA, K. & HIZUKURI, S., 1984. Examination of the purity and structure of amylose by gel-permeation chromatography. *Carbohydr. Res.* 132: 83-92.

TAKEDA, Y., TAKEDA, C., MIZUKAMI, H. & HANASHIRO, I., 1999. Structures of large, medium and small starch granules of barley grain. *Carbohydr. Polym.* 38: 109-114.

TANG, H., ANDO, H., WATANABE, K., TAKEDA, Y. & MITSUNAGA, T., 2000. Some physiochemical properties of small-, medium-, and large-granule starches in fractions of waxy barley grain. *Cereal Chem*. 77(1): 27-31.

TANG, H., ANDO, H., WATANABE, K., TAKEDA, Y. & MITSUNAGA, T., 2001. Physiochemical properties and structure of large, medium and small granule starches in fractions of normal barley endosperm. *Carbohydr. Res.* 330: 241-248.

TANG, H., MITSUNAGA, T. & KAWAMURA, Y., 2006. Molecular arrangement in blocklets and starch granule architecture. *Carbohydr. Polym.* 63: 555-560.

TANG, H., WATANABE, K. & MITSUNAGA, T., 2002. Structure and functionality of large, medium and small granule starches in normal and waxy barley endosperms. *Carbohydr. Polym.* 49: 217-224.

TATGE, H., MARSHALL, J., MARTIN, C., EDWARDS, E. & SMITH, A., 1999. Evidence that amylose synthesis occurs within the matrix of the starch granule in potato tubers. *Plant Cell Environ*. 22: 543-550.

TAYLOR, B.R., CRANSTOUN, D.A.S. & ROSCROW, J.C., 1993. The quality of winter wheat varieties for distilling from Scottish sites. *Aspects of Applied Biology* 36: 481-484.

TAYLOR, B.R. & ROSCROW, J.C., 1990. Factors affecting the quality of wheat grain for distilling in northern Scotland. *Aspects of Applied Biology* 25: 183-191.

TESTER, R.F., 1997a. Properties of damaged starch granules: composition and swelling properties of maize, rice, pea and potato starch fractions in water at various temperatures. *Food Hydrocolloids* 11: 293-301.

TESTER, R.F., 1997b. Starch: the polysaccharide fractions. In: Frazier, P.J., Donald, A.M., Richmond, P. (Eds.), *Starch: Structure and Functionality*. The Royal Society of Chemistry, London, UK, pp. 163-171.

TESTER, R.F. & KARKALAS, J., 2001. The effects of environmental conditions on the structural features and physico-chemical properties of starches. *Starch/Stärke* 53(4): 513-519.

TESTER, R.F., KARKALAS, J. & QI, X., 2004a. Starch – composition, fine structure and architecture. *J. Cereal Sci.* 39: 151-165.

TESTER, R.F., KARKALAS, J. & QI, X., 2004b. Starch structure and digestibility: Enzyme-Substrate relationship. *World's Poultry Science Journal* 60: 186-195.

TESTER, R.F. & MORRISON, W.R., 1990. Swelling and gelatinization of cereal starches. I. Effects of amylopectin, amylose and lipids. *Cereal Chem*. 67: 551.

TESTER, R.F., MORRISON, W.R., ELLIS, R.H., PIGOTT, J.R., BATTS, G.R., WHEELER, T.R., MORISON, J.I.L., HADLEY, P. & LEDWARD, D.A., 1995. Effects of elevated growth temperature and carbon dioxide levels on some physicochemical properties of wheat starch. *J. Cereal Sci*. 22: 63-71.

TESTER, R.F. & SOMMERVILLE, M.D., 2003. The effects of non-starch polysaccharides on the extent of gelatinisation, swelling and a-amylase hydrolysis of maize and wheat starches. *Food Hydrocolloids* 17: 41-54.

TESTER, R.F., SOUTH, J.B., MORRISON, W.R. & ELLIS, R.P., 1991. The effects of ambient temperature during the grain-filling period on the composition and properties of four barley genotypes. *J. Cereal Sci*. 13: 113-127.

TESTER, R.F., QI, X. & KARKALAS, J., 2006. Hydrolysis of native starches with amylases. *Anim. Feed Sci. Technol*. 130(1-2): 39-54.

TETLOW, I.J., 2006. Understanding storage starch biosynthesis in plants: a means to quality improvement. *Can. J. Bot*. 84: 1167-1185.

TETLOW, I.J., MORELL, M.K. & EMES, M.J., 2004. Recent developments in understanding the regulation of starch metabolism in higher plants. *J. Exp. Bot.* 55(406): 2131-2145.

TETLOW, I.J., WAIT, R., LU, Z.X., AKKASAENG, R., BOWSHER, C.G., ESPOSITO, S., KOSAR-HASHEMI, B., MORELL, M.K. & EMES, M.J., 2004. Protein phosphorylation in amyloplasts regulates starch branching enzyme activity and protein-protein interactions. *Plant Cell* 16: 694-708.

THEMEIER, H., HOLLMANN, J., NEESE, U. & LINDHAUER, M.G., 2005. Structural and morphological factors influencing the quantification of resistant starch II in starches of different botanical origin. *Carbohydr. Polym.* 61: 72-79.

THIEMT, E.M., SENN, T., OETTLER, G. & SCHÖN, C.C., 2006. Genetic variation for ethanol production in winter triticale. In: W.C. Botes, D. Boros, N. Darvey, P. Gustafson, R. Jessop, G.F. Marais, G: Oettler & D. Salmon (Eds.), *Proceedings of the 6th International Triticale Symposium*. 3-7 September 2006, Stellenbosch, South Africa, pp. 33.

THOMAS, S. & FELL, D.A., 1998. The role of multiple enzyme activation in metabolic flux control. *Advances in Enzyme Regulation* 38: 65-85.

THOMAS, K.C., HYNES, S.H. & INGLEDEW, W.M., 1996. Practical and theoretical considerations in the production of high concentrations of alcohol by fermentation. *Process Biochem*. 31: 321-331.

THOMAS, L., SENN, T. & PIEPER, H.J., 1991. Bioethanol aus Triticale, GIT. *Frachz. Lab*. 35: 1087-1089.

THOMPSON, D.B., 2000. On the non-random nature of amylopectin branching. *Carbohydr. Polym.* 43: 223-239.

THORBJØRNSEN, T., VILLAND, P., DENYER, K., OLSEN, O.A. & SMITH, A.M., 1996a. Distinct isoforms of ADPglucose pyrophosphorylase occur inside and outside the amyloplasts in barley endosperm. *Plant J.* 10: 243-250.

THORBJØRNSEN, T., VILLAND, P., KLECZKOWSKI, L.A. & OLSEN, O.A., 1996b. A single gene encodes two different transcripts for the ADP-glucose pyrophosphorylase small subunit from barley (*Hordeum vulgare*). *Biochem. J.* 313: 149-154.

TKACHENKO, N.M. & SERBIN, A.G., 1997. Botanika (in Ukrainian). Kharkiv, Osnova, 432 pp.

TOMLINSON, K. & DENYER, K., 2003. Starch synthesis in cereal grains. *Advances in Botanical Research* 40: 1-61.

TSVETKOV, S.M. & STOEVA, J., 2003. Bread Making Quality of Winter Hexaploid Triticale (×*Triticosecale* Wittmack) in Bulgaria. *Bulgarian Journal of Agriculturas Science* 9: 203-208.

TUFVESSON, F., SKRABANJA, V., BJORCK, I., ELMSTAHL, H.L. & ELIASSON, A.-C., 2001. Digestibility of starch systems containing amylose-glycerol monopalmitin complexes. *Lebensm. Wiss. u. Technol*. 34: 131-139.

TYMCHOUK, S.M., DIDENKO, S.YU. & RESHETNIKOV, M.V., 2002. Paralelizm genotypnoi minlyvosti kukurudzy ta gorokhu za fraktsijnym skladom krokhmalyu (in Ukrainian). *Visnyk kharkivs'kogo Nats. un-tu, Seriya Biologiya* 9(1): 45-51.

TYMCHOUK, S.M., RYABCHOUN, V.K., BOGUSLAVSKY, R.L., DIDENKO, S.YU. & GERASIMOV, N.V., 2001. Collection of maize endosperm structure mutants: working out, evaluation and utilization in breeding. *Genetic Collections, Isogenic and Alloplasmic Lines*. Int. Conf., Novosibirsk, pp. 240-242.

TYYNELA, J. & SCHULMAN, A.H., 1993. An analysis of soluble starch synthase isozymes from the developing grains of normal and shx cv. Bomi barley (*Hordeum vulgare*). *Physiol. Plant* 89: 835-841.

TYYNELA, J., STITT, M., LONNEBORG, A., SMEEKENS, S. & SCHULMAN, A.H., 1995. Metabolism of starch synthesis in developing grains of the *shx* shrunken mutant of barley (*Hordeum vulgare*). *Physiol. Plant* 93: 77-84.

TZIOTIS, A., SEETHARAMAN, K., WONG, K.S., KLUCINEC, J.D., JANE, J.L. & WHITE, P.J., 2004. Structural properties of starch fractions isolated from normal and mutant corn genotypes using different methods. *Cereal Chem.* 81(5): 611-620.

UMEMOTO, T., NAKAMURA, Y. & ISHIKURA, N., 1995. Activity of starch synthase and the amylose content in rice endosperm. *Phytochemistry* 40: 1613-1616.

UMEMOTO, T., YANO, M., SATOH, H., SHOMURA, A. & NAKAMURA, Y., 2002. Mapping of a gene responsible for the difference in amylopectin structure between japonica-type and indica-type rice varieties. *Theor. Appl. Genet.* 104: 1-8.

UTHAYAKUMARAN, S., BATEY, I.L. & WRIGLEY, C.W., 2005. On-the-spot identification of grain variety and wheat-quality type by Lab-on-a-chip capillary electrophoresis. *J. Cereal Sci.* 41: 371-374.

VAN EYS, J.E., OFFNER, A. & BACH, A., 2004. Chemical Analysis. Manual of quality analyses for soybean products in the feed industry. *American Soybean Association Europe*. Retrieved November 4, 2007, from http://www.asa-europe.org/Library/library_e.htm

VAN WEY, L., 2009. Social and distributional impacts of biofuel production. In: R.W. Howarth & S. Bringezu (Eds.), *Biofuels: Environmental Consequences and Interactions with Changing Land Use*, pp. 197-206. Proceedings of the Scientific Committee on Problems of the Environment (SCOPE) International Biofuels Project Rapid Assessment, 22-25 September 2008, Gummersbach, Germany. Cornell University, Ithaca NY, USA. Retrieved February 28, 2009, from http://cip.cornell.edu/biofuels/

VANDEPUTTE, G.E. & DELCOUR, J.A., 2004. From sucrose to starch granule to starch physical behaviour: a focus on rice starch. *Carbohydr. Polym.* 58: 245-266.

VARUGHESE, G., PFEIFFER, W.H. & PENA, R.J., 1997. Triticale: A reappraisal. Retrieved November 4, 2007, from http://www.worldbank.org/html/cgiar/newsletter/april97/8tritic.html

VASANTHAN, T. & BHATTY, R.S., 1996. Physicochemical properties of small- and large-granule starches of waxy, regular and high amylose barleys. *Cereal Chem*. 73: 199-207.

VASIL, V., CASTILLO, A.M., FROMM, M.E. & VASIL, I.K., 1992. Herbicide Resistant Fertile Transgenic Wheat Plants Obtained by Microprojectile Bombardment of Regenerable Embryogeni Callus. *Bio Technology* 10: 667-674.

VELASCO, L., GOFFMAN, F.D. & BECKER, H.C., 1999. Development of calibration equations to predict oil content and fatty acid composition in Brassicaceae germplasm by near-infrared reflectance spectroscopy. *Journal of the American Oil Chemists' Society* 76: 25-30.

VELASCO, L. & MOLLERS, C., 2002. Nondestructive assessment of protein content in single seeds of rapeseed (*Brassica napus* L.) by near-infrared reflectance spectroscopy. *Euphytica* 123: 89-93.

VERMEYLEN, R., DERYCKE, V., DELCOUR, J.A., GODERIS, B., REYNAERS, H. & KOCH, M.H.J., 2006. Structural transformations during gelatinization of starches in limited water: combined wide- and small-angle x-ray scattering study. *Biomacromolecules* 7: 1231-1238.

VERMEYLEN, R., GODERIS, B., REYNAERS, H. & DELCOUR, J.A., 2004. Amylopectin molecular structure reflected in macromolecular organization of granular starch. *Biomacromolecules* 5: 1775-1786.

VIGNAUX, N., DOEHLERT, D.C., HEGSTAD, J., ELIAS, E.M., MCMULLEN, M.S., GRANT, L.A. & KIANIAN, S.F., 2004. Grain Quality Characteristics and Milling Performance of Full and Partial Waxy Durum Lines. *Cereal Chem*. 81(3): 377-383.

VISSER, R.G.F., SOMHORST, I., KUIPERS, G.J., RUYS, N.J., FEENSTRA, W.J. & JACOBSEN, E., 1991. Inhibition of expression of the gene for granule-bound starch synthase in potato by antisens constructs. *Mol. Gen*. 225: 289-296.

VOGEL, K.P. & JUNG, H.-J.G., 2001. Genetic Modification of Herbaceous Plants for Feed and Fuel. *Bioenergy* 20(1): 15-49.

VUAUROVIA, V.M. & PEJIN, D.J., 2007. Production of bioethanol from triticale. *Zbornik Matice srpske za prirodne nauke / Proc. Nat. Sci. Matica Srpska Novi Sad* 113: 285-291.

WAL, M., D'HULT, C., VINCKEN, J.P., BULLEON, A., VISSER, R. & BALL, S., 1998. Amylose is synthesizes *in vitro* by extension of and cleavage from amylopectin. *J. Biol. Chem*. 273: 22232-22240.

WANG, T.L., BOGRACHEVA, T.YA. & HEDLEY, C.L., 1998. Starch: as simple as A, B, C? *J. Exp. Bot.* 49(320): 481-502.

WANG, T.L. & HEDLEY, C.L., 1993. Genetic and developmental analysis of the seed. In: R. Casey, D.R. Davies (Eds.), *Peas: Genetics, Molecular Biology and Biotechnology*. Cambrige: CAB Int., pp. 83-120.

WANG, P., SINGH, V., XUE, H., JOHNSTON, D.B., *et al.*, 2007. Comparison of raw starch hydrolyzing enzyme with conventional liquefaction and saccharification enzymes in dry-grind corn processing. *Cereal Chem.* 84(1): 10-14.

WANG, S., THOMAS, K.C., SOSULSKI, K., INGLEDEW, W.M. & SOSULSKI, F.W., 1999. Grain pearling and very high gravity (VHG) fermentation technologies for fuel alcohol production from rye and triticale. *Process Biochemistry* 34: 421-428.

WANG, Y.J., WHITE, P., POLLAK, L. & JANE, J., 1993. Characterization of starch structure of 17 maize endosperm mutant genotypes with Oh43 inbred line background. *Cereal Chem*. 70(2): 171-179.

WANG, Z.Y., WU, Z.L., XING, Y.Y., ZHENG, F.G., GUO, X.L., ZHANG, W.G. & HONG, M.M., 1990. Nucleotide sequence of rice waxy gene. *Nucleot. Acids Res*. 18: 5898.

WATANABE, N., NODA, Y., GOTO, N., MIURA, H., 1998. Variation for apparent amylose content of endosperm starch in *Triticum durum* and *Aegilops sqarrosa*. *Euphytica* 101(2): 283-286.

WATKINS, M.S., HAMMOCK, B.D., NEWMAN, J.W. & GERMAN, J.B., 2001. Individual metabolism should guide agriculture towards foods for improved health and nutrition. *American Journal of Clinical Nutrition* 74: 283-286.

WEBER, C.J., HAUGAARD, V., FESTERSEN, R. & BERTELSEN, G., 2002. Production and applications of biobased packaging materials for the food industry. *Food Addit. Contam*. 19(Suppl.): 172-177.

WEBER, H., HEIM, U., GOLOMBEK, S., BORISJUK, L., MATEUFFEL, R. & WOBUS, U., 1998. Expression of a yeast-derived invertase in developing cotyledons of *Vicia narbonensis* alters the carbohydrate state and affects storage functions. *Plant J*. 16: 163-172.

WEIGHTMAN, R.M., FOULKES, J.F., ALAVA, J., GREENWELL, P. & SNAPE, J., 2005. Physiological traits influencing hardness and vitreosity in wheat grain. *Final Report on Project AR0908* to the UK Department for Environment, Food and Rural Affairs (DEFRA), UK. Retrieved November 4, 2007, from http://randd.defra.gov.uk/Document.aspx?Document=AR0908_6641_FRP.doc

WELLIE-STEPHAN, O., 2005. Breeding of winter wheat varieties for bio-ethanol production / Zuchtung von Winterweizensorten fur die Bioethanolproduktion. *Getreide Magazin* (2): 141-143.

WESLEY, S.V., HELLIWELL, C., SMITH, N.A., WANG, M.-B., ROUSE, D., LIU, Q., GOODING, P., SINGH, S., ABBOTT, D., STOUTJESDIJK, P., ROBINSON, S., GLEAVE, A., GREEN, A. & WATERHOUSE, P.M., 2001b. Constructs for efficient, effective and high throughput gene silencing in plants. *Plant J.* 27: 581-590.

WESLEY, I.J., LARROQUE, O., OSBORNE, B.G., AZUDIN, N., ALLEN, H. & SKERRITT, J.H., 2001a. Measurement of gliadin and glutenin content of flour by NIR spectroscopy. *J. Cereal Sci.* 34: 125-133.

WHITE, P., 1994. Properties of corn starch. In: Hallauer, A.R. (Ed.), *Specialty Corns.* Boca Raton, Fl.: CRC Press Inc., pp. 29-54.

WHITE, M.A., 1998. The chemistry behind carbonless copy paper. *J. Chemical Educ.* 75: 1119-1120.

WHITT, S.R., WILSON, L.M., TENAILLON, M.I., GAUT, B.S. & BUCKLER, E.S., 2002. Genetic diversity and selection in maize starch pathway. *Proc. New-York Acad. Sci*. 99(20): 12959-12962.

WICKRAMASINGHE, H.A.M. & MIURA, H., 2003. Gene dosage effect of the wheat Wx alleles and their interaction on amylose synthesis in the endosperm. *Euphytica* 132: 303-310.

WILLIAMS, P.C., 2001. Implementation of near-infrared reflectance technology. In: P. Williams, K. Norris (Eds.), *Near-infrared Technology in the Agricultural and Food Industries*. AACC International: St. Paul, MN, USA, pp. 145-169.

WILLIAMS, P. & NORRIS, K., 1987. Near-Infrared Technology in the Agriculture and Food Industries. American Association of Cereal Chemists, St. Paul, MN, USA. 330 pp.

WILLIAMS, P. & NORRIS, K. (Eds.), 2001. Near-Infrared Technology in the Agricultural and Food Industries. 2nd ed., American Association of Cereal Chemists, St. Paul, MN, USA. 145 pp.

WILLIAMS, P.C. & SOBERING, D.C., 1996. How do we do it: a brief summary of the methods we use in developing near infrared calibrations. In: Davies, A.M.C., Williams, P.C. (Eds.), *Near Infrared Spectroscopy: The Future Waves*. NIR Publications, Chichester, UK. pp. 185-188.

WOLD, S., SJÖSTRÖM, M. & ERIKSSON, L., 2001. PLS-regression: a basic tool of chemometrics. *Chemometrics and Intelligent Laboratory Systems* 58: 109-130.

WOODROW, I.E. & BERRY, J.A., 1988. Enzymatic regulation of photosynthetic CO2 fixation. *Annu. Rev. Plant Physiol. Plant Mol. Biol*. 39: 533-594.

WOODS, P.J., WEISZ, J. & MAHN, W., 1991. Molecular characteristics of cereal β-glucans. II. Size exclusion chromatography for comparison of molecular weights. *Cereal Chem*. 68: 530.

WU, Y.V., 1989. Protein-Rich Residue from Ethanolic Fermentation of High-Lysine, Dent, Waxy, and White Corn Varieties. *Cereal Chem*. 66(6): 506-509.

WU, J.G. & SHI, C.H., 2004. Prediction of grain weight, brown rice weight and amylose content in single rice grains using near-infrared reflectance spectroscopy. *Field Crop Research* 87: 13-21.

WU, J.G., SHI, C.H. & ZHANG, X.M., 2002. Estimating the amino acid composition in milled rice by near-infrared reflectance spectroscopy. *Field Crops Research* 75: 1-7.

WU, X., ZHAO, R., WANG, D. & BEAN, S.R., 2006b. Ethanol Production as Affected by Sorghum Chemical Composition, Chemical Structure and Physical Properties. *2006 ASAE Annual Meeting*, Paper number 067076. Retrieved November 4, 2007, from http://asae.frymulti.com/abstract.asp?aid=21034&t=2

WU, X., ZHAO, R., WANG, D., BEAN, S.R., SEIB, P.A., TUINSTRA, M.R., CAMPBELL, M. & O'BRIEN, A., 2006. Effects of amylase, corn protein, and corn fiber contents on production of ethanol from starch-rich media. *Cereal Chem*. 83(5): 569-575.

XICCATO, G., TROCINO, A., DE BOEVER, J.L., MAERTENS, L., CARABANO, R., PASCUAL, J.J., PEREZ, J.M., GIDENNE, T. & FALCAO-E-CUNHA, L., 2003. Prediction of chemical composition, nutritive value and ingredient composition of European compound feeds for rabbits by near infrared reflectance spectroscopy (NIRS). *Animal Feed Science and Technology* 104: 153-168.

XU, Q., WANG, L., NEWMAN, R.K., NEWMAN, C.W. & GRAHAM, H., 1997. Influence of the hulless, waxy starch and short-awn genes on the composition of barleys. *J. Cereal Sci*. 26: 251-257.

XUE, Q., NEWMAN, R.K. & NEWMAN, C.W., 1996. Effects of heat treatment of barley starches on in vitro digestibility and glucose responses in rats. *Cereal Chem*. 73: 588-592.

YAMAMORI, M., FUJITA, S., HAYAKAWA, K., MATSUKI, J. & YASUI, T., 2000. Genetic elimination of starch granule protein, SGP-1, of wheat generates and altered starch with apparent high amylase. *Theor. Appl. Genet.* 101: 21-29.

YAMAMORI, M., NAKAMURA, T., ENDO, T.R. & NAGAMINE, T., 1994. Waxy Protein Deficiency and Chromosomal Locations of Coding Genes in Common Wheat. *Theor. Appl. Genet*. 89: 179-184.

YAMAMORI, M. & QUYNH, N.T., 2000. Differential effects of Wx-A1, -B1 and -D1 protein deficiencies on apparent amylose content and starch pasting properties in common wheat. *Theor. Appl. Genet*. 100: 32-38.

YAN, L., BHAVE, M., FAIRCLOUGH, R., KONIK, C., RAHMAN, S. & APPELS, R., 2000. The genes encoding granule-bound starch synthases at the waxy loci of A, B and D progenitors of wheat. *Genome* 43: 264-272.

YAN, W. & HUNT, L.A., 1998. Genotype by environment interaction and crop yield. *Plant breeding reviews* 16: 135-178.

YANAGISAWA, T., KIRIBUCHI-OTOBE, C. & FUJITA, M., 2004. Increase in Apparent Amylose Content and Change in Starch Pasting Properties at Cool Growth Temperatures in Mutant Wheat. *Cereal Chem.* 81(1): 26-30.

YANAGISAWA, T., KIRIBUCHI-OTOBE, C. & YOSHIDA, H., 2001. An alanine to threonine change in the Wx-D1 protein reduces GBSS I activity in waxy mutant wheat. *Euphytica* 121: 209-214.

YAO, Y., THOMPSON, D.B. & GUILTINAN, M.J., 2004. Maize starch-branching enzyme isoforms and amylopectin structure. In the absence of starch branching enzyme IIb, the further absence of starch-branching enzyme Ia leads to increased branching. *Plant Physiol.* 136: 3515-3523.

YASUI, T., MATSUKI, J., SASAKI, T. & YAMAMORI, M., 1996. Amylose and lipid contents, amylopectin structure, and gelatinization properties of waxy wheat (*Triticum aestivum* L.) starch. *J. Cereal Sci.* 24: 131-137.

YASUI, T., MATSUKI, J., SASAKI, T. & YAMAMORI, M., 1997. Waxy endosperm mutants of bread wheat (*Triticum aestivum* L.) and their starch properties. *Breed. Sci.* 47: 161-163.

YASUI, T., SASAKI, T. & MATSUKI, J., 1999. Milling and flour pasting properties of waxy endosperm mutant lines of bread wheat (*Triticum aestivum* L). *J. Sci. Food. Agric*. 79: 687-692.

YOO, S. & JANE, J., 2002a. Structural and physical characteristics of waxy and other wheat starches. *Carbohydr. Polym*. 49: 297-305.

YOO, S. & JANE, J., 2002b. Molecular weights and gyration radii of amylopectins determined by high-performance size-exclusion chromatography equipped with multi-angle laser light scattering and refractive index detection. *Carbohydr. Polym*. 49: 307-314.

YOOSIN, S. & SORAPIPATANA, C.A., 2007. Study of ethanol production cost for gasoline substitution in Thailand and its competitiveness. *Thammasat. Int. J. Sci. Technol.* 12: 69-80.

YOSHIDA, M., FUJII, M., NIKUNI, Z. & MARUO, B., 1958. The appositive growth of starch granules in beans as revealed by autoradiography. *Bull. Agric. Chem. Soc. Jpn*. 22: 127.

YOSHIDA, H., NOZAKI, K., HANASHIRO, I., YAGI, F., ITO, H., HONMA, M., MATSUI, H. & TAKEDA, Y., 2003. Structure and physico-chemical properties of starches from kidney bean seeds at immature, premature and mature stages of development. *Carbohydr. Res*. 338: 463-469.

YU, A.S-M., 2003. Transgenic rice expressing a thermostable amylopullulanase in seeds leads to starch autohydrolysis and production of high-protein flour. *American Society of Plant Biologists Meeting*: 2003 July 25-30, Honlulu, Hawaii. Retrieved November 4, 2007, from http://abstracts.aspb.org/pb2003/public/P34/0355.html

YUAN, C.K.R., THOMPSON, D.B. & BOYER, C.D., 1993. Fine structure of amylopectin in relation to gelatinization and retrogradation behaviour in maize starches from three *wx*-containing genotypes in two inbred lines. *Cereal Chem*. 70: 81-89.

YURYEV, V.P., KRIVANDIN, A.V., KISELEVA, V.I., WASSERMAN, L.A., GENKINA, N.K., FORNAL, J., BLASZCZAK, W. & SCHIRALDI, A., 2004. Structural parameters of amylopectin clusters and semi-crystalline growth rings in wheat starches with different amylose content. *Carbohydr. Res.* 339(16): 2683-2691.

ZEEMAN, S.C., SMITH, S.M. & SMITH, A.M., 2002. The priming of amylose synthesis in Arabidopsis leaves. *Plant Physiol*. 128: 1069-1076.

ZEEMAN, S.C., UMEMOTO, T., LUE, W.L., AU-YEUNG, P., MARTIN, C., SMITH, A.M. & CHEN, J., 1998. A mutant of *Arabidopsis* lacking a chloroplastic isoamylase accumulates both starch and phytoglycogen. *Plant Cell* 10: 1699-1711.

ZHAN, X., WANG, D., TUINSTRA, M.R., BEAN, S., SEIB, P.A. & SUN, X.S., 2003. Ethanol and lactic acid production as affected by sorghum genotype and location. *Industrial Crops and Products* 18: 245-255.

ZHANG, G., CHEN, J., WANG, J. & DING, S., 2001. Cultivar and environmental effects on (1-3, 1-4)-b-d-glucan and protein content in malting barley. *J. Cereal Sci*. 34, 295-301.

ZHANG, X.L., COLLEONI, C., RATUSHNA, V., SIRGHLE-COLLEONI, M., JAMES, M.G. & MYERS, A.M., 2004. Molecular characterization demonstrates that the Zea mays gene sugary2 codes for the starch synthase isoform SSIIa. *Plant Molecular Biology* 54: 865-869.

ZHANG, G. & HAMAKER, B.R., 1998. Low a-amylase starch digestibility of cooked sorghum flours and the effect of protein. *Cereal Chem.* 75: 710-713.

ZHAO, X.C., BATEY, I.L., SHARP, P.J., CROSBIE, G., BARCLAY, I., WILSON, R., MORELL, M.K. & APPELS, R., 1998. A single genetic locus associated with starch granule properties and noodle quality in wheat. *J. Cereal Sci*. 27: 7-13.

ZHUSHMAN, A.I., KARPOV, V.G., BAKULINA, L.F. & BURAVLEVA, T.N., 2001. Razrabotka tekhnologii dieticheskikh produktov pitaniya na osnove krakhmala (in Russian). In: *Krakhmal i krakhmalsoderzhashchie istochniki – struktura, svojstva i novye tekhnologii:* Sbornik dokladov mezhdunarodnoj konferentsii. Moscow, p. 160.

ZIEGLER, G.R., THOMPSON, D.B. & CASASNOVAS, J., 1993. Dynamic measurement of starch granule swelling. *Cereal Chem*. 70: 247-251.

ZILLINSKY, F.J. & BORLAUG, N.E., 1971. Progress in developing triticale as an economic crop. *CIMMYT Res. Bull*. No. 17, Mexico. 27 pp.

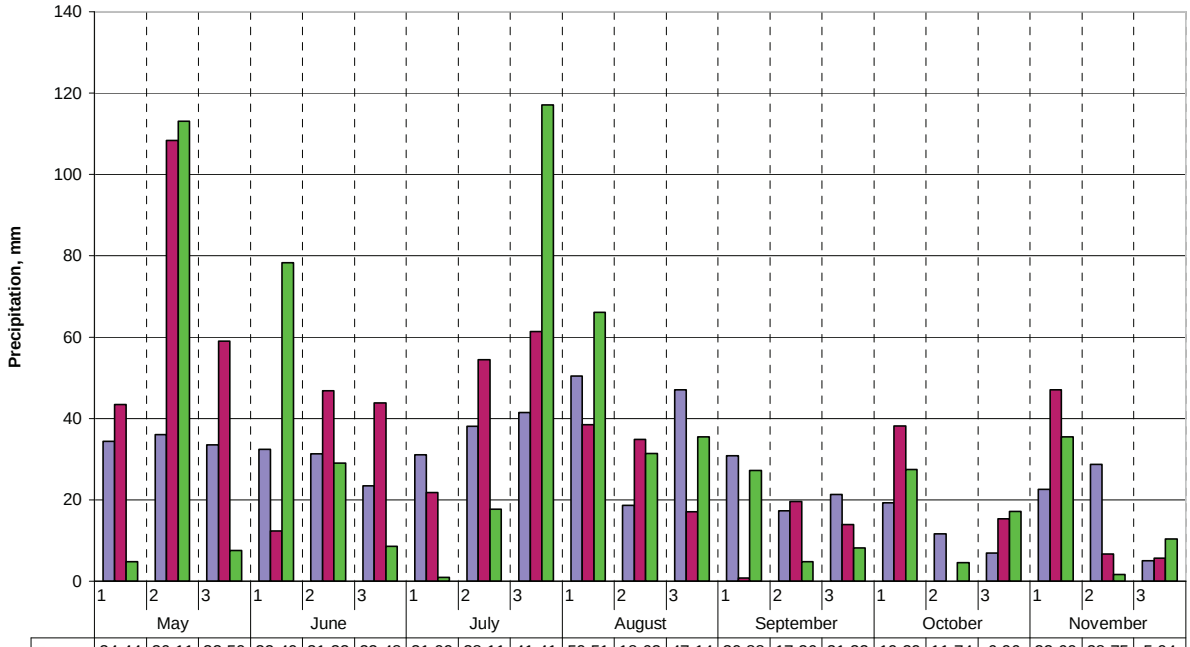
ZOBEL, H.F., 1988a. Starch crystal transformations and their industrial importance. *Starch/Stärke* 40: 1-7.

ZOBEL, H.F., 1988b. Molecules to granules: A comprehensive starch review. *Starch/Stärke* 44: 44-50.

ZUGENMAIER, P., 2003. Symmetry and packing consideration of crystalline amylosics and their solvent complexes. *XI Int. Starch Convent*., Moscow, p. 32.

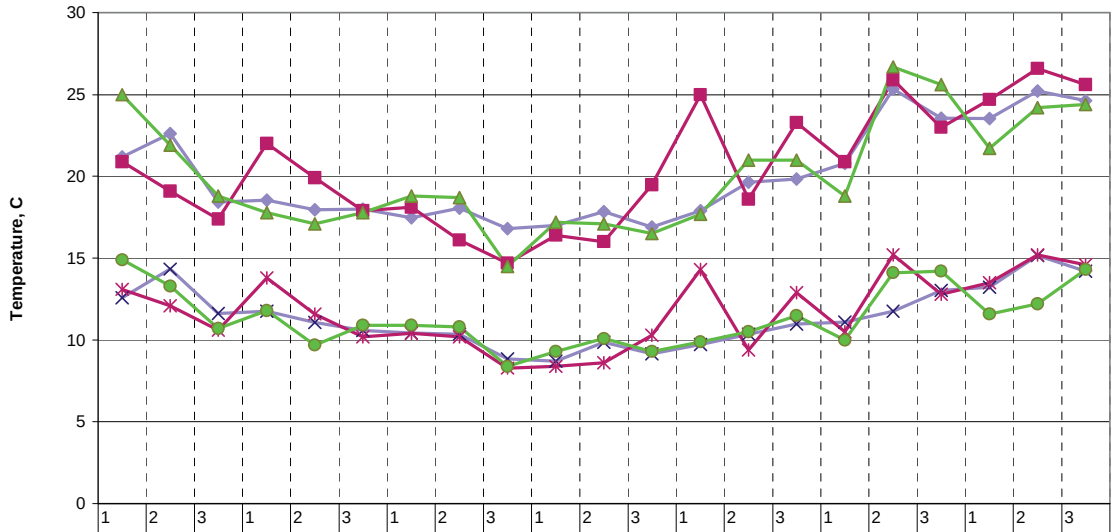
Addendum 1: The 2006 and 2007 season's 10-day average and long-term (LT) average data for precipitation, maximum and minimum temperature for each trial location

Figure A4.1.1. Mariendahl 2006 and 2007 season's 10-day average precipitation with 9 years average (LT) data



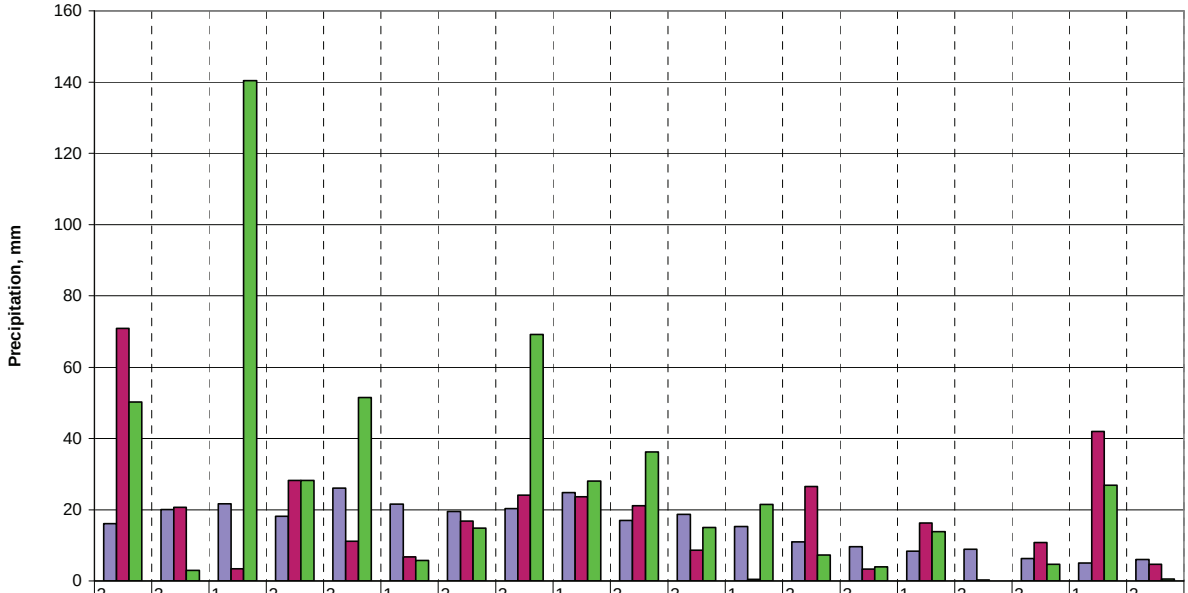
	May			June			July			August			September			October			November		
LT	34.44	36.11	33.50	32.40	31.33	23.48	31.09	38.11	41.41	50.51	18.63	47.14	30.88	17.36	21.32	19.29	11.74	6.96	22.60	28.75	5.04
2006	43.40	108.4	59.00	12.40	46.80	43.80	21.80	54.40	61.40	38.50	34.90	17.10	0.80	19.60	14.00	38.20	0.00	15.40	47.10	6.70	5.70
2007	4.90	113.0	7.50	78.30	29.10	8.60	1.00	17.70	117.1	66.10	31.40	35.50	27.30	4.90	8.20	27.50	4.60	17.20	35.50	1.70	10.40

Figure A4.1.2. Mariendahl 2006 and 2007 season's 10-day average maximum and minimum temperatures with 11 years average (LT) data



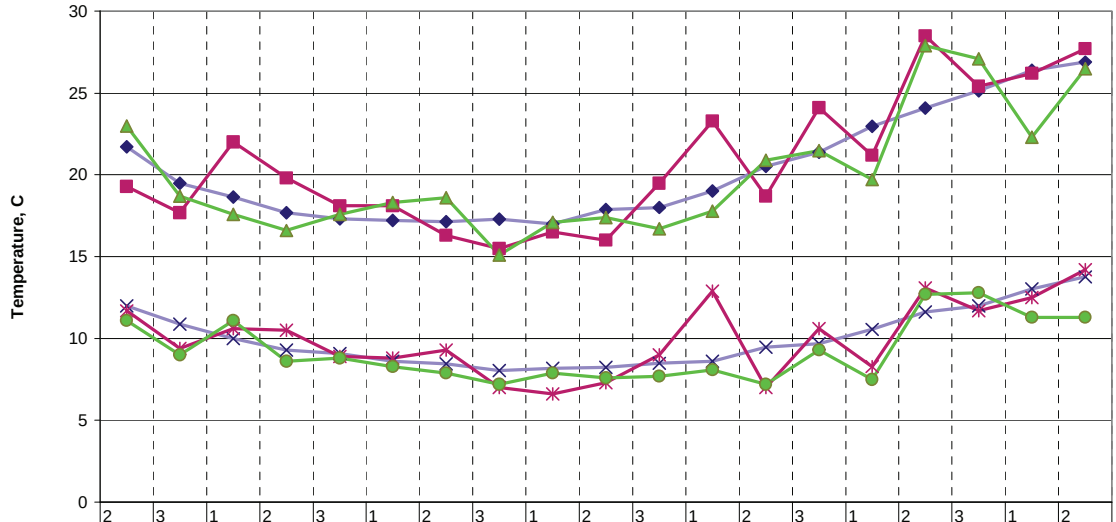
	May			June			July			August			September			October			November		
Tmax, LT	21.19	22.62	18.43	18.55	17.97	18.01	17.47	18.05	16.81	17.01	17.83	16.91	17.91	19.64	19.84	20.80	25.34	23.56	23.54	25.21	24.61
Tmax, 2006	20.90	19.10	17.40	22.00	19.90	17.90	18.10	16.10	14.70	16.40	16.00	19.50	25.00	18.60	23.30	20.90	25.90	23.00	24.70	26.60	25.60
Tmax, 2007	25.00	21.90	18.80	17.80	17.10	17.80	18.80	18.70	14.50	17.20	17.10	16.50	17.70	21.00	21.00	18.80	26.70	25.60	21.70	24.20	24.40
Tmin, LT	12.58	14.34	11.61	11.78	11.07	10.58	10.43	10.37	8.86	8.72	9.86	9.16	9.71	10.35	10.98	11.10	11.78	13.03	13.23	15.17	14.20
Tmin, 2006	13.10	12.10	10.60	13.80	11.60	10.20	10.40	10.20	8.30	8.40	8.60	10.30	14.30	9.40	12.90	10.50	15.20	12.80	13.50	15.20	14.60
Tmin, 2007	14.90	13.30	10.70	11.80	9.70	10.90	10.90	10.80	8.40	9.30	10.10	9.30	9.90	10.50	11.50	10.00	14.10	14.20	11.60	12.20	14.30

Figure 4.1.3. Langgewens 2006 and 2007 season's 10-day average precipitation with 41 years average (LT) data



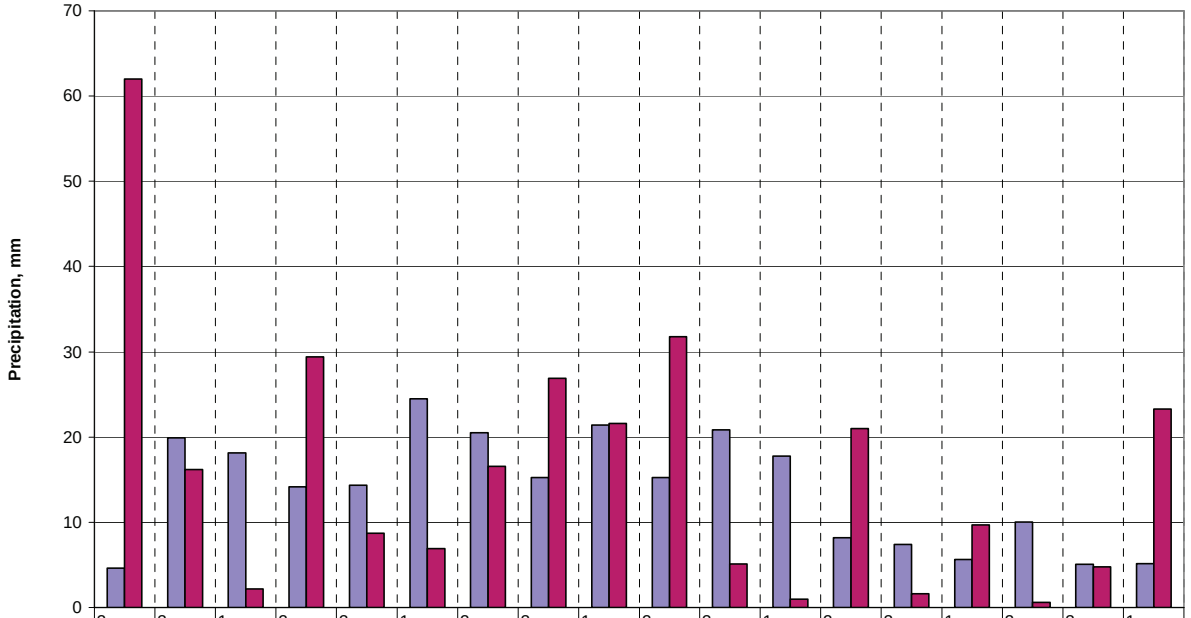
	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2
	May		June			July			August			September			October			November	
LT	16.14	20.02	21.62	18.08	26.03	21.50	19.51	20.31	24.81	16.95	18.65	15.34	11.04	9.67	8.42	8.89	6.28	5.04	6.00
2006	70.80	20.60	3.40	28.20	11.10	6.70	16.80	24.00	23.60	21.10	8.60	0.40	26.50	3.30	16.20	0.20	10.80	41.90	4.60
2007	50.20	3.00	140.40	28.20	51.50	5.80	14.90	69.10	28.00	36.10	15.00	21.40	7.20	4.00	13.80	0.00	4.70	26.80	0.60

Figure 4.1.4. Langgewens 2006 and 2007 season's 10-day average maximum and minimum temperatures with 41 years average (LT) data



	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2
			June			July			August			September			October			November	
Tmax, LT	21.72	19.50	18.64	17.69	17.33	17.21	17.15	17.29	17.00	17.89	18.01	19.01	20.52	21.38	22.96	24.09	25.13	26.41	26.90
Tmax, 2006	19.30	17.70	22.00	19.80	18.10	18.10	16.30	15.50	16.50	16.00	19.50	23.30	18.70	24.10	21.20	28.50	25.40	26.20	27.70
Tmax, 2007	23.00	18.70	17.60	16.60	17.60	18.30	18.60	15.10	17.10	17.40	16.70	17.80	20.90	21.50	19.70	27.90	27.10	22.30	26.50
Tmin, LT	11.99	10.89	9.99	9.31	9.07	8.62	8.45	8.05	8.20	8.23	8.48	8.61	9.48	9.69	10.55	11.62	11.99	13.02	13.76
Tmin, 2006	11.70	9.40	10.60	10.50	8.90	8.80	9.30	7.00	6.60	7.30	9.00	12.90	7.00	10.60	8.30	13.10	11.70	12.50	14.20
Tmin, 2007	11.10	9.00	11.10	8.60	8.80	8.30	7.90	7.20	7.90	7.60	7.70	8.10	7.20	9.30	7.50	12.70	12.80	11.30	11.30

Figure 4.1.5. Vredenburg 2006 season's 10-day average precipitation with 10 years average (LT) data

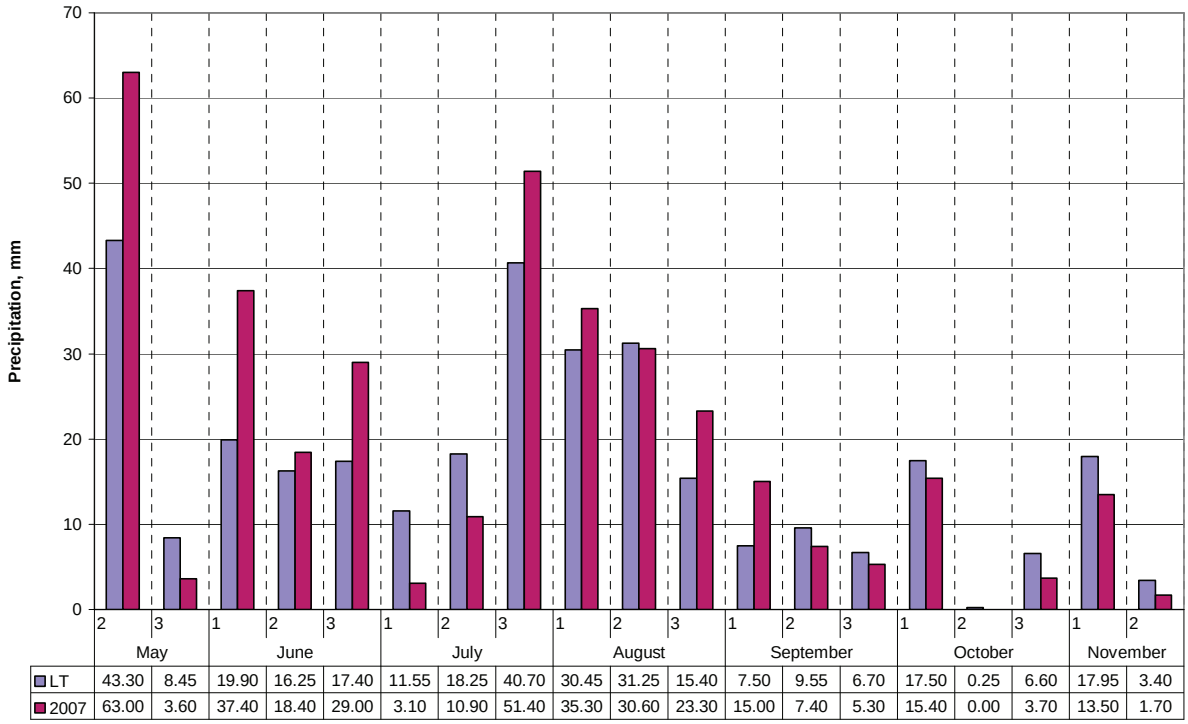


LT	4.65	19.87	18.13	14.14	14.34	24.50	20.52	15.26	21.42	15.26	20.83	17.75	8.20	7.41	5.63	10.05	5.06	5.15
2006	62.00	16.20	2.20	29.40	8.70	6.90	16.60	26.90	21.60	31.80	5.10	1.00	21.00	1.60	9.70	0.60	4.80	23.30

Figure 4.1.6. Vredenburg 2006 season's 10-day average maximum and minimum temperatures with 9 years average (LT) data

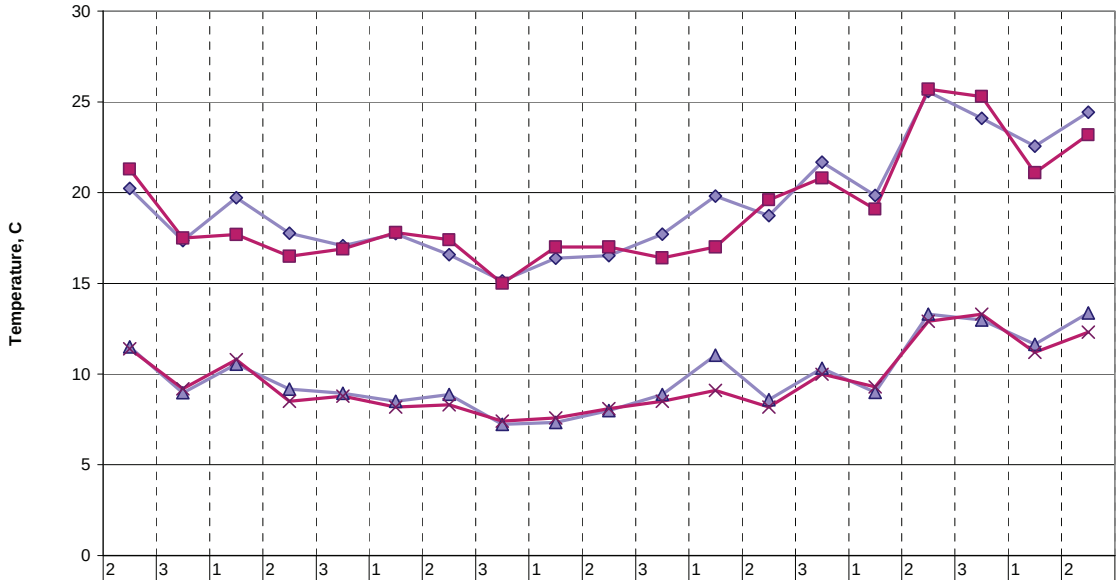
Tmax, LT	25.61	20.93	20.11	19.33	18.99	18.93	18.55	18.25	18.21	19.34	19.38	19.70	22.66	22.61	24.03	26.32	26.35	26.63
Tmax, 2006	20.40	18.80	23.20	20.50	19.20	19.60	17.40	16.90	17.70	17.20	21.10	24.30	20.40	24.90	22.20	29.70	26.30	27.60
Tmin, LT	10.43	8.90	7.42	7.46	6.43	6.45	5.98	5.82	6.63	6.57	7.13	7.45	8.04	7.97	8.84	10.96	10.39	11.21
Tmin, 2006	10.00	6.90	6.00	8.40	6.80	5.90	8.50	6.60	5.70	6.40	7.30	10.00	6.10	8.20	5.60	11.30	10.00	10.70

Figure 4.1.7. Klipheuwel 2007 season's 10-day average precipitation with 3 years average (LT) data



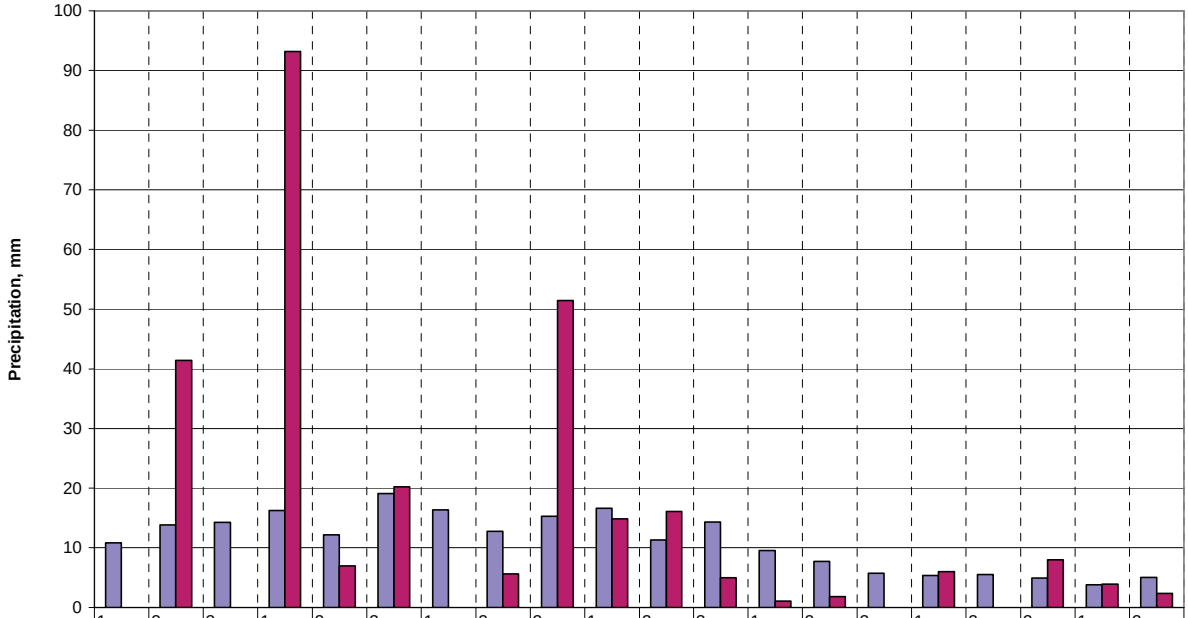
	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2
	May		June			July			August			September			October			November	
LT	43.30	8.45	19.90	16.25	17.40	11.55	18.25	40.70	30.45	31.25	15.40	7.50	9.55	6.70	17.50	0.25	6.60	17.95	3.40
2007	63.00	3.60	37.40	18.40	29.00	3.10	10.90	51.40	35.30	30.60	23.30	15.00	7.40	5.30	15.40	0.00	3.70	13.50	1.70

Figure 4.1.8. Klipheuwel 2007 season's 10-day average maximum and minimum temperatures with 3 years average (LT) data



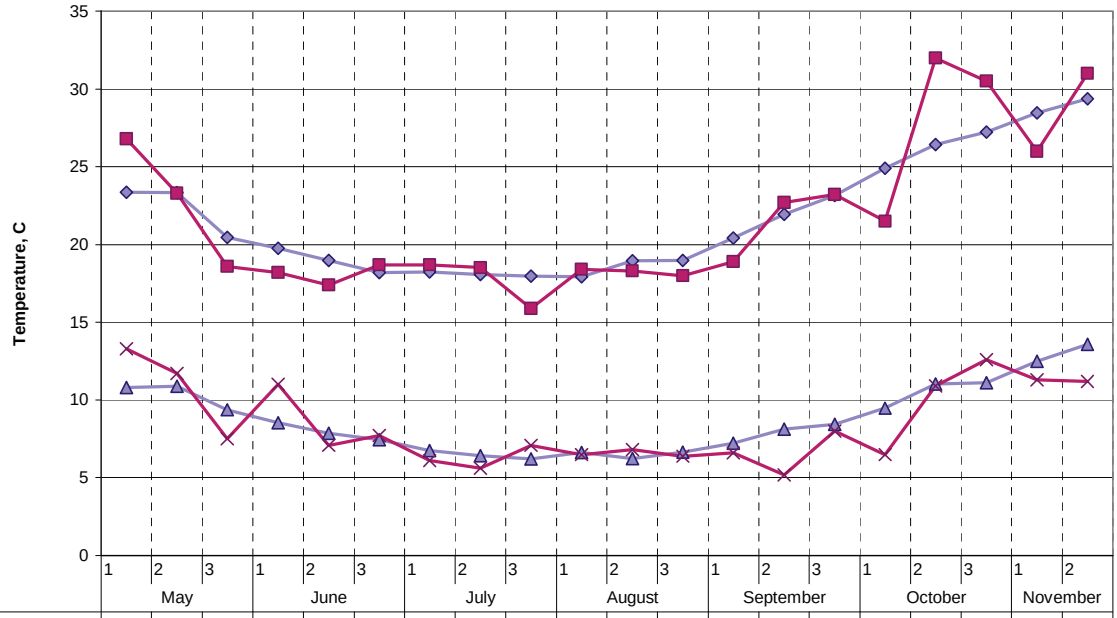
	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2
			June			July			August			September			October			November	
Tmax, LT	20.22	17.36	19.71	17.77	17.06	17.74	16.58	15.13	16.38	16.54	17.72	19.80	18.75	21.67	19.85	25.58	24.10	22.56	24.44
Tmax, 2007	21.30	17.50	17.70	16.50	16.90	17.80	17.40	15.00	17.00	17.00	16.40	17.00	19.60	20.80	19.10	25.70	25.30	21.10	23.20
Tmin, LT	11.50	8.97	10.56	9.17	8.94	8.50	8.88	7.23	7.35	8.00	8.87	11.03	8.60	10.33	8.98	13.30	12.98	11.64	13.37
Tmin, 2007	11.40	9.20	10.80	8.50	8.80	8.20	8.30	7.40	7.60	8.10	8.50	9.10	8.20	10.00	9.30	12.90	13.30	11.20	12.30

Figure 4.1.9. Piketberg 2007 season's 10-day average precipitation with 37 years average (LT) data



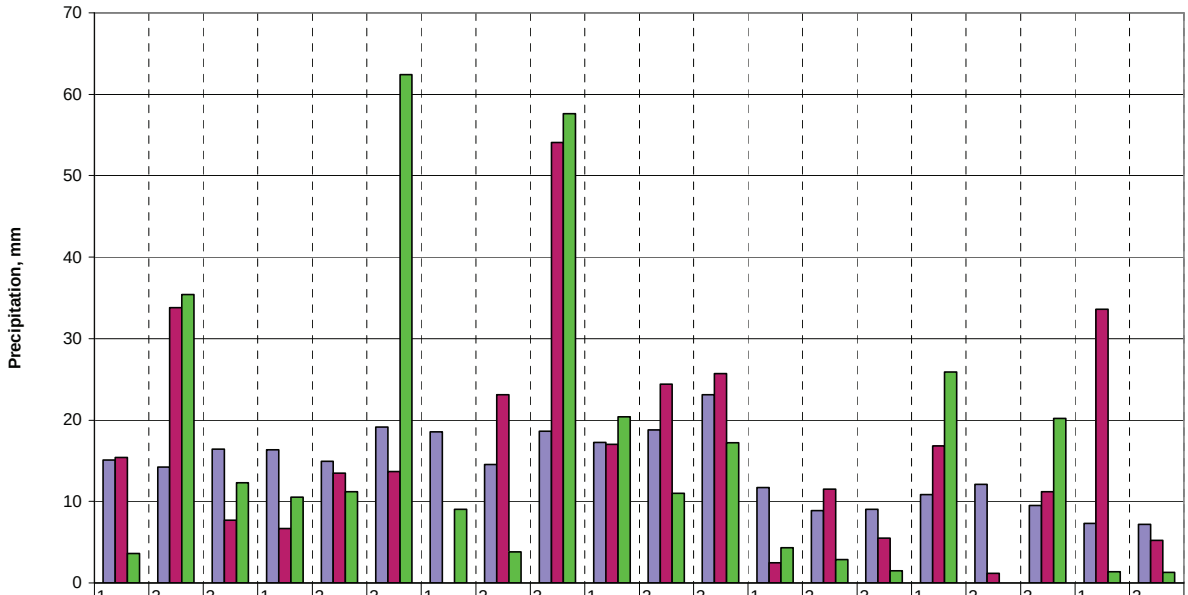
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2
	May			June			July			August			September			October			November	
LT	10.85	13.86	14.30	16.27	12.20	19.07	16.37	12.77	15.29	16.67	11.28	14.33	9.54	7.72	5.72	5.34	5.52	4.95	3.82	5.08
2007	0.00	41.40	0.00	93.20	7.00	20.20	0.00	5.60	51.40	14.90	16.10	5.00	1.10	1.80	0.00	6.00	0.00	8.00	3.90	2.40

Figure 4.1.10. Piketberg 2007 season's 10-day average maximum and minimum temperatures with 37 years average (LT) data



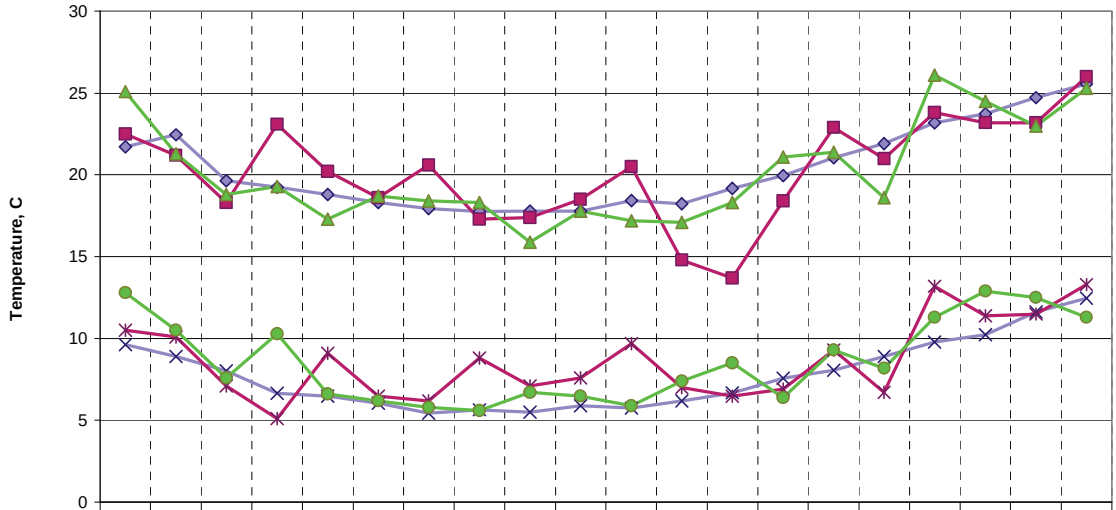
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2
	May			June			July			August			September			October			November	
Tmax, LT	23.36	23.33	20.46	19.75	18.99	18.20	18.23	18.06	17.96	17.91	18.96	18.97	20.41	21.94	23.15	24.91	26.44	27.24	28.45	29.37
Tmax, 2007	26.80	23.30	18.60	18.20	17.40	18.70	18.70	18.50	15.90	18.40	18.30	18.00	18.90	22.70	23.20	21.50	32.00	30.50	26.00	31.00
Tmin, LT	10.79	10.87	9.37	8.55	7.87	7.46	6.75	6.41	6.20	6.63	6.24	6.65	7.23	8.13	8.45	9.48	11.03	11.12	12.48	13.58
Tmin, 2007	13.30	11.70	7.50	11.00	7.10	7.70	6.10	5.60	7.10	6.50	6.80	6.40	6.60	5.20	8.00	6.50	10.90	12.60	11.30	11.20

Figure 4.1.11. Roodebloem 2006 and 2007 season's 10-day average precipitation with 46 years average (LT) data



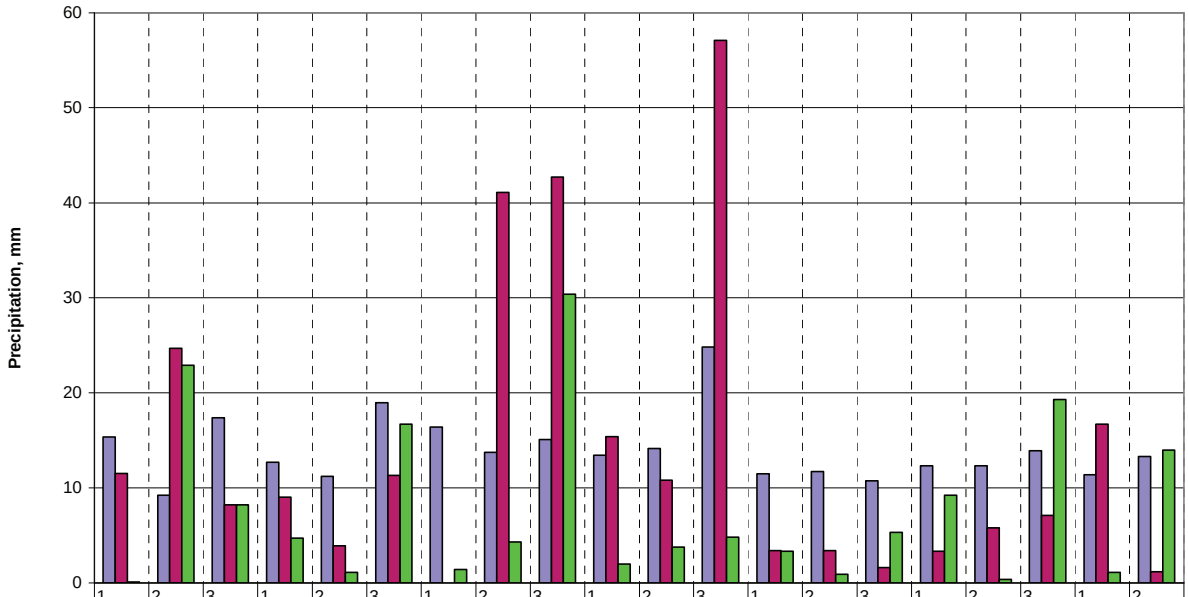
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2
	May			June			July			August			September			October			November	
LT	15.07	14.24	16.39	16.37	14.90	19.16	18.54	14.54	18.65	17.27	18.78	23.08	11.71	8.86	9.00	10.87	12.11	9.50	7.29	7.19
2006	15.40	33.80	7.70	6.70	13.50	13.70	0.00	23.10	54.10	17.00	24.40	25.70	2.50	11.50	5.50	16.80	1.20	11.20	33.60	5.20
2007	3.60	35.40	12.30	10.50	11.20	62.40	9.00	3.80	57.60	20.40	11.00	17.20	4.30	2.90	1.50	25.90	0.00	20.20	1.40	1.30

Figure 4.1.12. Roodebloem 2006 and 2007 season's 10-day average maximum and minimum temperatures with 34 years average (LT) data



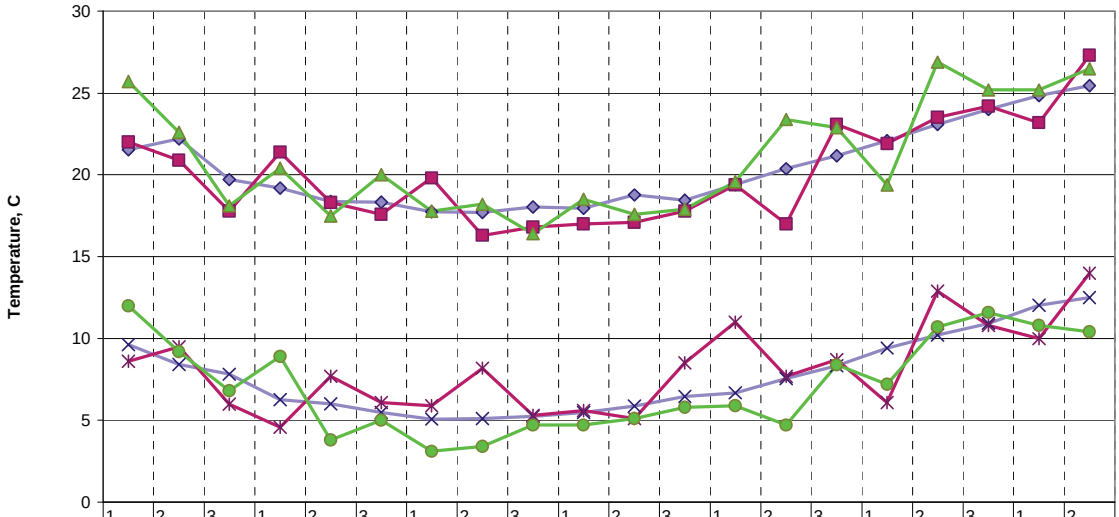
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2
	May			June			July			August			September			October			November	
Tmax, LT	21.70	22.44	19.65	19.24	18.80	18.31	17.93	17.76	17.78	17.79	18.44	18.23	19.17	19.95	21.05	21.91	23.19	23.73	24.73	25.57
Tmax, 2006	22.50	21.20	18.30	23.10	20.20	18.60	20.60	17.30	17.40	18.50	20.50	14.80	13.70	18.40	22.90	21.00	23.80	23.20	23.20	26.00
Tmax, 2007	25.10	21.30	18.80	19.30	17.30	18.70	18.40	18.30	15.90	17.80	17.20	17.10	18.30	21.10	21.40	18.60	26.10	24.50	23.00	25.30
Tmin, LT	9.62	8.90	8.02	6.66	6.50	6.04	5.46	5.64	5.49	5.89	5.75	6.20	6.68	7.58	8.07	8.90	9.80	10.24	11.62	12.46
Tmin, 2006	10.50	10.10	7.10	5.10	9.10	6.50	6.20	8.80	7.10	7.60	9.70	7.00	6.50	6.90	9.30	6.70	13.20	11.40	11.50	13.30
Tmin, 2007	12.80	10.50	7.60	10.30	6.60	6.20	5.80	5.60	6.70	6.50	5.90	7.40	8.50	6.40	9.30	8.20	11.30	12.90	12.50	11.30

Figure 4.1.13. Tygerhoek 2006 and 2007 season's 10-day average precipitation with 38 years average (LT) data



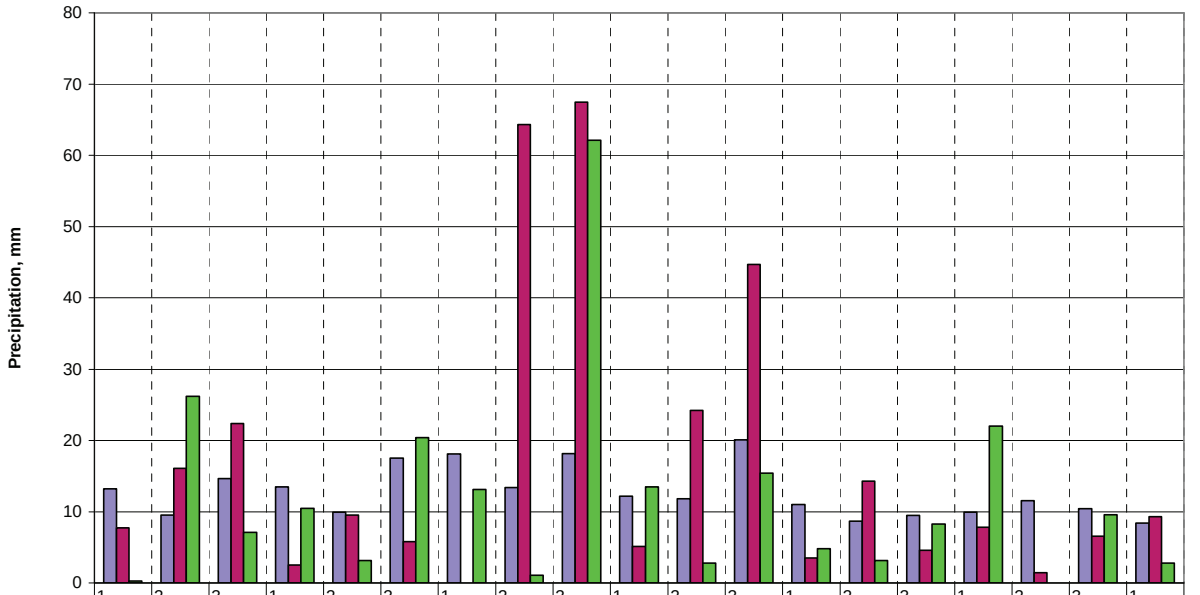
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2
	May			June			July			August			September			October			November	
LT	15.32	9.23	17.39	12.71	11.21	18.94	16.39	13.71	15.09	13.42	14.13	24.80	11.47	11.73	10.74	12.34	12.32	13.91	11.37	13.30
2006	11.50	24.70	8.20	9.00	3.90	11.30	0.00	41.10	42.70	15.40	10.80	57.10	3.40	3.40	1.60	3.30	5.80	7.10	16.70	1.20
2007	0.10	22.90	8.20	4.70	1.10	16.70	1.40	4.30	30.40	2.00	3.80	4.80	3.30	0.90	5.30	9.20	0.40	19.30	1.10	14.00

Figure 4.1.14. Tygerhoek 2006 and 2007 season's 10-day average maximum and minimum temperatures with 36 years average (LT) data



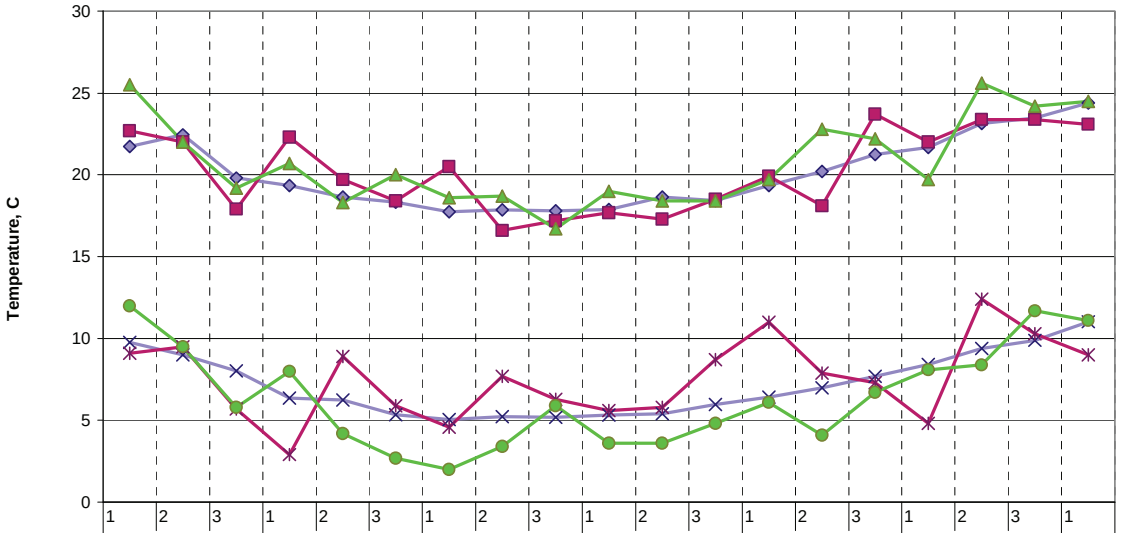
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2
	May			June			July			August			September			October			November	
Tmax, LT	21.53	22.21	19.70	19.19	18.37	18.32	17.73	17.71	18.03	17.95	18.78	18.46	19.42	20.37	21.17	22.09	23.10	24.00	24.84	25.47
Tmax, 2006	22.00	20.90	17.80	21.40	18.30	17.60	19.80	16.30	16.80	17.00	17.10	17.80	19.40	17.00	23.10	21.90	23.50	24.20	23.20	27.30
Tmax, 2007	25.70	22.60	18.10	20.40	17.50	20.00	17.80	18.20	16.40	18.50	17.60	17.90	19.60	23.40	22.90	19.40	26.90	25.20	25.20	26.50
Tmin, LT	9.63	8.42	7.82	6.27	6.03	5.47	5.08	5.10	5.26	5.48	5.86	6.47	6.68	7.54	8.35	9.43	10.22	10.93	12.05	12.50
Tmin, 2006	8.60	9.50	6.00	4.60	7.70	6.10	5.90	8.20	5.30	5.60	5.10	8.50	11.00	7.70	8.70	6.10	12.90	10.80	10.00	14.00
Tmin, 2007	12.00	9.20	6.80	8.90	3.80	5.00	3.10	3.40	4.70	4.70	5.10	5.80	5.90	4.70	8.40	7.20	10.70	11.60	10.80	10.40

Figure 4.1.15. Napier 2006 and 2007 season's 10-day average precipitation with 35 years average (LT) data



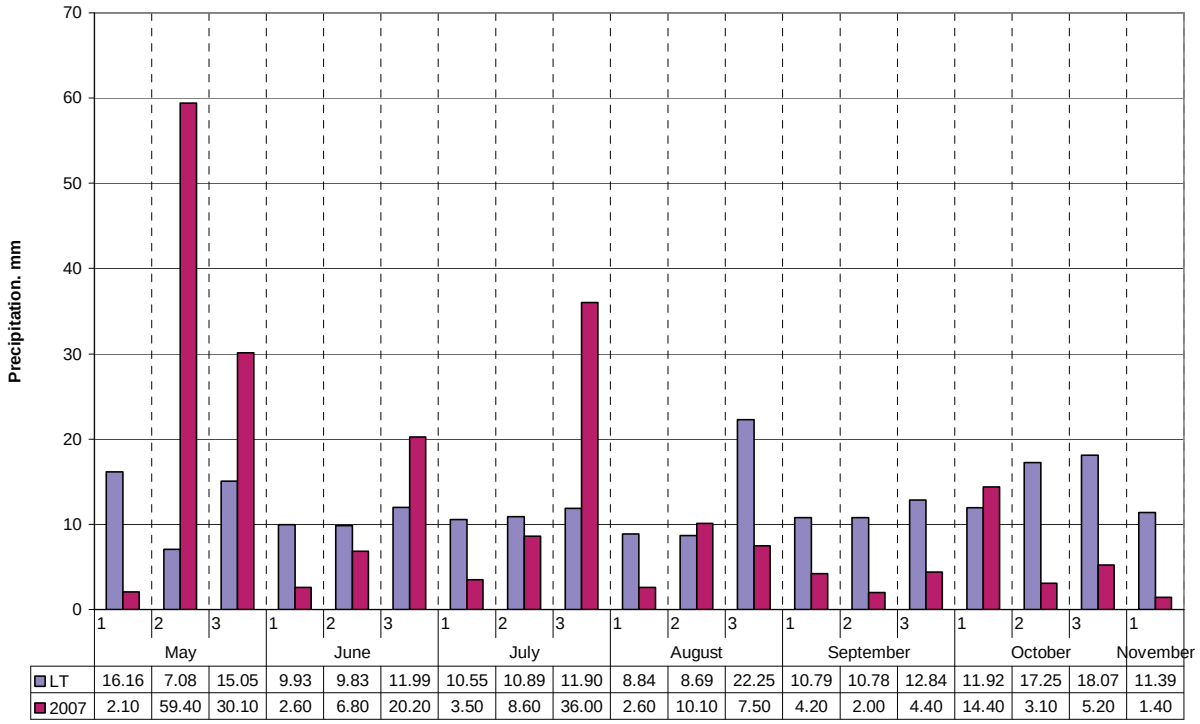
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1
	May			June			July			August			September			October			November
LT	13.18	9.53	14.64	13.46	9.92	17.48	18.09	13.36	18.11	12.16	11.76	20.06	11.01	8.65	9.46	9.92	11.58	10.44	8.36
2006	7.70	16.10	22.40	2.50	9.50	5.80	0.00	64.30	67.50	5.10	24.20	44.70	3.50	14.30	4.60	7.80	1.40	6.60	9.30
2007	0.30	26.20	7.10	10.50	3.10	20.40	13.10	1.10	62.10	13.50	2.80	15.40	4.80	3.10	8.30	22.00	0.00	9.60	2.80

Figure 4.1.16. Napier 2006 and 2007 season's 10-day average maximum and minimum temperatures with 35 years average (LT) data



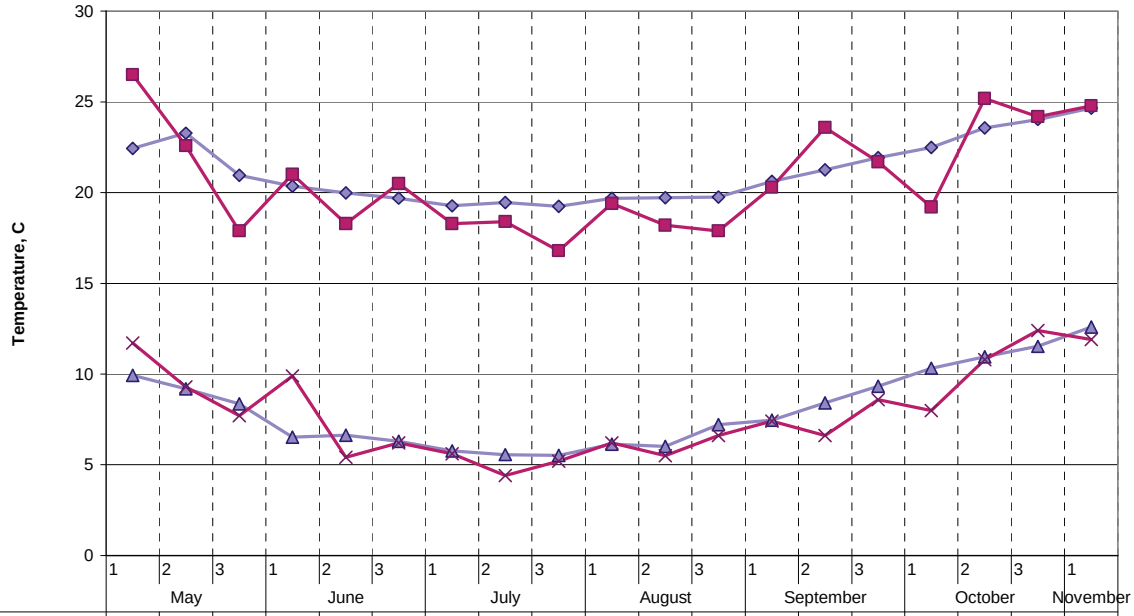
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1
	May			June			July			August			September			October			November
Tmax, LT	21.73	22.45	19.82	19.33	18.65	18.33	17.75	17.86	17.81	17.88	18.64	18.43	19.34	20.21	21.24	21.69	23.15	23.49	24.41
Tmax, 2006	22.70	22.00	17.90	22.30	19.70	18.40	20.50	16.60	17.20	17.70	17.30	18.50	19.90	18.10	23.70	22.00	23.40	23.40	23.10
Tmax, 2007	25.50	22.00	19.20	20.70	18.30	20.00	18.60	18.70	16.70	19.00	18.40	18.40	19.70	22.80	22.20	19.70	25.60	24.20	24.50
Tmin, LT	9.77	9.00	8.02	6.36	6.24	5.36	5.05	5.23	5.19	5.32	5.40	5.98	6.42	6.99	7.69	8.41	9.41	9.89	11.04
Tmin, 2006	9.10	9.50	5.70	2.90	8.90	5.90	4.60	7.70	6.30	5.60	5.80	8.70	11.00	7.90	7.30	4.80	12.40	10.30	9.00
Tmin, 2007	12.00	9.50	5.80	8.00	4.20	2.70	2.00	3.40	5.90	3.60	3.60	4.80	6.10	4.10	6.70	8.10	8.40	11.70	11.10

Figure 4.1.17. Riversdale 2007 season's 10-day average precipitation with 33 years average (LT) data



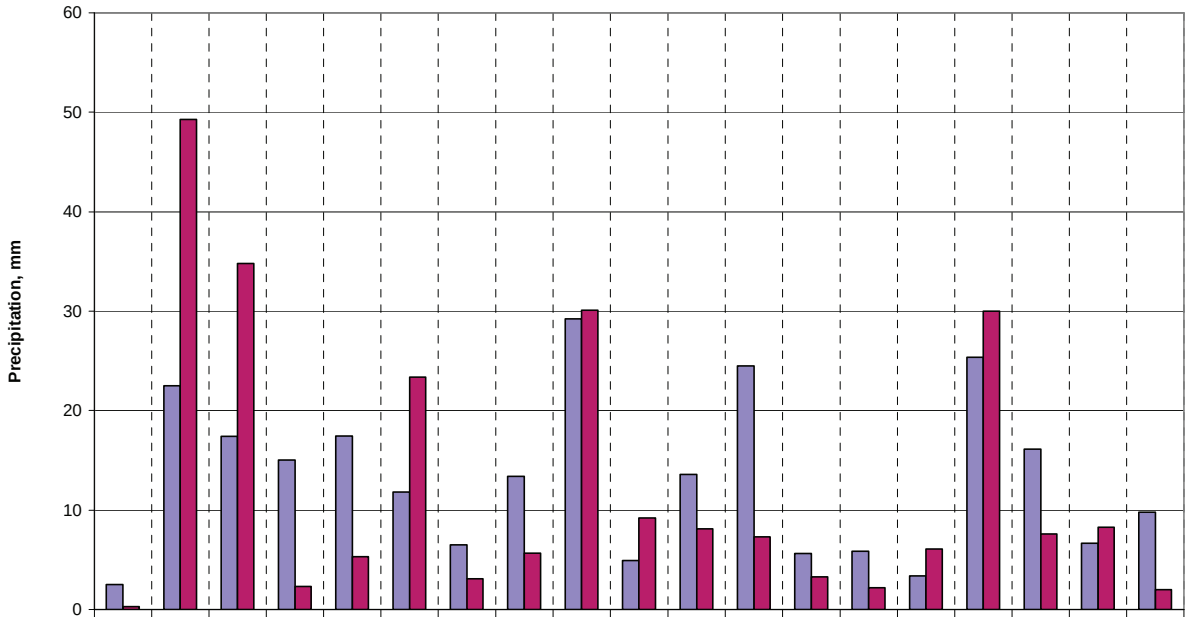
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1
	May			June			July			August			September			October			November
LT	16.16	7.08	15.05	9.93	9.83	11.99	10.55	10.89	11.90	8.84	8.69	22.25	10.79	10.78	12.84	11.92	17.25	18.07	11.39
2007	2.10	59.40	30.10	2.60	6.80	20.20	3.50	8.60	36.00	2.60	10.10	7.50	4.20	2.00	4.40	14.40	3.10	5.20	1.40

Figure 4.1.18. Riversdale 2007 season's 10-day average maximum and minimum temperatures with 32 years average (LT) data



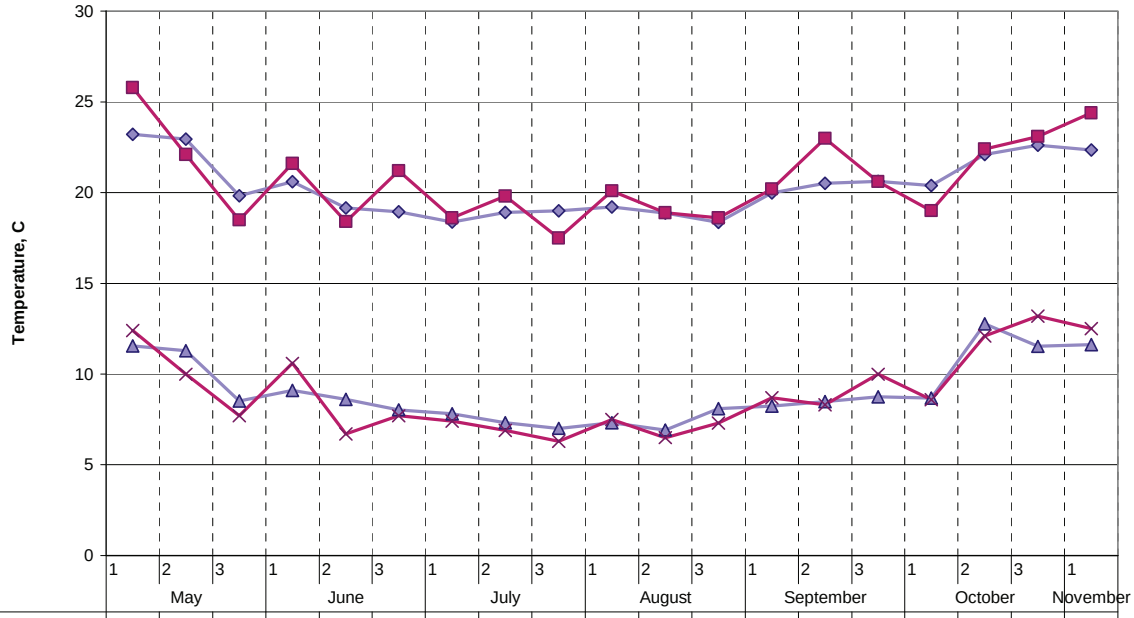
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1
	May			June			July			August			September			October			November
Tmax, LT	22.44	23.27	20.96	20.37	19.99	19.70	19.28	19.46	19.26	19.70	19.71	19.77	20.62	21.25	21.92	22.51	23.56	24.04	24.65
Tmax, 2007	26.50	22.60	17.90	21.00	18.30	20.50	18.30	18.40	16.80	19.40	18.20	17.90	20.30	23.60	21.70	19.20	25.20	24.20	24.80
Tmin, LT	9.92	9.20	8.37	6.53	6.63	6.29	5.76	5.56	5.51	6.15	6.00	7.22	7.45	8.41	9.32	10.33	10.94	11.53	12.60
Tmin, 2007	11.70	9.30	7.70	9.90	5.40	6.20	5.60	4.40	5.20	6.20	5.50	6.60	7.40	6.60	8.60	8.00	10.80	12.40	11.90

Figure 4.1.19. Albertinia 2007 season's 10-day average precipitation with 3 years average (LT) data



	May			June			July			August			September			October			November
LT	2.53	22.50	17.40	15.03	17.47	11.83	6.50	13.40	29.23	4.90	13.60	24.48	5.65	5.85	3.38	25.38	16.10	6.65	9.80
2007	0.30	49.30	34.80	2.30	5.30	23.40	3.10	5.70	30.10	9.20	8.10	7.30	3.30	2.20	6.10	30.00	7.60	8.30	2.00

Figure 4.1.20. Albertinia 2007 season's 10-day average maximum and minimum temperatures with 3 years average (LT) data



	May			June			July			August			September			October			November
Tmax, LT	23.21	22.95	19.84	20.61	19.17	18.94	18.38	18.91	19.00	19.21	18.87	18.37	19.98	20.53	20.63	20.38	22.09	22.62	22.35
Tmax, 2007	25.80	22.10	18.50	21.60	18.40	21.20	18.60	19.80	17.50	20.10	18.90	18.60	20.20	23.00	20.60	19.00	22.40	23.10	24.40
Tmin, LT	11.55	11.28	8.52	9.11	8.62	8.04	7.81	7.33	7.02	7.32	6.93	8.09	8.24	8.47	8.74	8.69	12.78	11.53	11.61
Tmin, 2007	12.40	10.00	7.70	10.60	6.70	7.70	7.40	6.90	6.30	7.50	6.50	7.30	8.70	8.30	10.00	8.60	12.10	13.20	12.50

Addendum 2: Plant height, days from planting to heading, grain yield and test weight raw data, the 2006 and 2007 season trials

Table A4.4.1. Mariendahl 2006 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	101	100	70	2881.20
2	1	2	**CB**	119	110	68	1683.64
3	1	3	**CC**	100	105	68	2179.52
4	1	4	**CD**	84	100	74	1956.08
5	1	5	**CE**	98	125	72	2910.60
6	1	6	**D1**	104	105	70	2263.80
7	1	7	**DB**	102	120	72	2367.68
8	1	8	**YA**	101	110	74	2581.32
9	1	9	**DC**	104	105	70	2794.96
10	1	10	**YB**	99	95	74	2087.40
11	1	11	**YC**	104	95	70	2120.72
12	1	12	**DD**	100	110	74	2261.84
13	1	13	**DE**	103	105	70	2902.76
14	1	14	**DF**	95	110	72	2879.24
15	1	15	**DG**	108	115	70	2024.68
16	1	16	**DH**	101	115	72	2785.16
17	1	17	**Y1**	101	115	70	3045.84
18	1	18	**Y2**	102	120	72	3353.56
19	1	19	**YD**	101	115	72	2257.92
20	1	20	**Y3**	102	120	72	2924.32
21	2	1	**CA**	.	.	.	3555.44
22	2	2	**CB**	.	.	.	2571.52
23	2	3	**CC**	.	.	.	2926.28
24	2	4	**CD**	.	.	.	2469.60
25	2	5	**CE**	.	.	.	2463.72
26	2	6	**D1**	.	.	.	3771.04
27	2	7	**DB**	.	.	.	2861.60
28	2	8	**YA**	.	.	.	2444.12
29	2	9	**DC**	.	.	.	3000.76
30	2	10	**YB**	.	.	.	2626.40
31	2	11	**YC**	.	.	.	1789.48
32	2	12	**DD**	.	.	.	2706.76
33	2	13	**DE**	.	.	.	2892.96
34	2	14	**DF**	.	.	.	2520.56
35	2	15	**DG**	.	.	.	2600.92
36	2	16	**DH**	.	.	.	2440.20
37	2	17	**Y1**	.	.	.	2989.00
38	2	18	**Y2**	.	.	.	2479.40
39	2	19	**YD**	.	.	.	2555.84
40	2	20	**Y3**	.	.	.	3324.16

Table A4.4.1. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	.	2898.84
42	3	2	**CB**	.	.	.	2148.16
43	3	3	**CC**	.	.	.	2287.32
44	3	4	**CD**	.	.	.	1930.60
45	3	5	**CE**	.	.	.	3449.60
46	3	6	**D1**	.	.	.	2187.36
47	3	7	**DB**	.	.	.	1958.04
48	3	8	**YA**	.	.	.	2838.08
49	3	9	**DC**	.	.	.	3330.04
50	3	10	**YB**	.	.	.	2683.24
51	3	11	**YC**	.	.	.	3188.92
52	3	12	**DD**	.	.	.	2410.80
53	3	13	**DE**	.	.	.	3482.92
54	3	14	**DF**	.	.	.	2663.64
55	3	15	**DG**	.	.	.	2018.80
56	3	16	**DH**	.	.	.	2536.24
57	3	17	**Y1**	.	.	.	3071.32
58	3	18	**Y2**	.	.	.	2994.88
59	3	19	**YD**	.	.	.	3104.64
60	3	20	**Y3**	.	.	.	3034.08
61	4	1	**CA**	.	.	.	2887.08
62	4	2	**CB**	.	.	.	1617.00
63	4	3	**CC**	.	.	.	2783.20
64	4	4	**CD**	.	.	.	1928.64
65	4	5	**CE**	.	.	.	2928.24
66	4	6	**D1**	.	.	.	3141.88
67	4	7	**DB**	.	.	.	2781.24
68	4	8	**YA**	.	.	.	2977.24
69	4	9	**DC**	.	.	.	3114.44
70	4	10	**YB**	.	.	.	2397.08
71	4	11	**YC**	.	.	.	3214.40
72	4	12	**DD**	.	.	.	2767.52
73	4	13	**DE**	.	.	.	2945.88
74	4	14	**DF**	.	.	.	2987.04
75	4	15	**DG**	.	.	.	2516.64
76	4	16	**DH**	.	.	.	3330.04
77	4	17	**Y1**	.	.	.	3061.52
78	4	18	**Y2**	.	.	.	2857.68
79	4	19	**YD**	.	.	.	3126.20
80	4	20	**Y3**	.	.	.	3969.00

* Codified due to confidentiality.

Table A4.4.2. Langgewens 2006 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	97	105	70	2759.68
2	1	2	**CB**	100	115	70	1189.72
3	1	3	**CC**	96	120	66	2802.80
4	1	4	**CD**	81	110	72	3441.76
5	1	5	**CE**	95	125	69	2377.48
6	1	6	**D1**	97	110	70	1716.96
7	1	7	**DB**	97	130	66	1965.88
8	1	8	**YA**	96	125	74	2465.68
9	1	9	**DC**	99	110	72	2024.68
10	1	10	**YB**	99	110	74	1901.20
11	1	11	**YC**	99	115	72	2297.12
12	1	12	**DD**	100	125	72	1313.20
13	1	13	**DE**	100	110	70	2303.00
14	1	14	**DF**	93	125	72	1703.24
15	1	15	**DG**	102	125	72	1950.20
16	1	16	**DH**	96	120	72	3598.56
17	1	17	**Y1**	99	120	74	3853.36
18	1	18	**Y2**	101	120	74	2714.60
19	1	19	**YD**	96	125	70	3202.64
20	1	20	**Y3**	99	125	72	3506.44
21	2	1	**CA**	.	.	.	2326.52
22	2	2	**CB**	.	.	.	680.12
23	2	3	**CC**	.	.	.	1215.20
24	2	4	**CD**	.	.	.	2614.64
25	2	5	**CE**	.	.	.	2455.88
26	2	6	**D1**	.	.	.	1581.72
27	2	7	**DB**	.	.	.	2171.68
28	2	8	**YA**	.	.	.	2953.72
29	2	9	**DC**	.	.	.	4490.36
30	2	10	**YB**	.	.	.	2540.16
31	2	11	**YC**	.	.	.	2838.08
32	2	12	**DD**	.	.	.	1632.68
33	2	13	**DE**	.	.	.	2093.28
34	2	14	**DF**	.	.	.	980.00
35	2	15	**DG**	.	.	.	1436.68
36	2	16	**DH**	.	.	.	1511.16
37	2	17	**Y1**	.	.	.	2742.04
38	2	18	**Y2**	.	.	.	2363.76
39	2	19	**YD**	.	.	.	3396.68
40	2	20	**Y3**	.	.	.	1816.92

Table A4.4.2. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	.	4776.52
42	3	2	**CB**	.	.	.	3298.68
43	3	3	**CC**	.	.	.	3776.92
44	3	4	**CD**	.	.	.	3426.08
45	3	5	**CE**	.	.	.	3947.44
46	3	6	**D1**	.	.	.	3328.08
47	3	7	**DB**	.	.	.	4049.36
48	3	8	**YA**	.	.	.	3829.84
49	3	9	**DC**	.	.	.	5474.28
50	3	10	**YB**	.	.	.	4523.68
51	3	11	**YC**	.	.	.	3255.56
52	3	12	**DD**	.	.	.	3592.68
53	3	13	**DE**	.	.	.	4088.56
54	3	14	**DF**	.	.	.	4278.68
55	3	15	**DG**	.	.	.	4170.88
56	3	16	**DH**	.	.	.	3804.36
57	3	17	**Y1**	.	.	.	3537.80
58	3	18	**Y2**	.	.	.	4466.84
59	3	19	**YD**	.	.	.	4284.56
60	3	20	**Y3**	.	.	.	3767.12
61	4	1	**CA**	.	.	.	3908.24
62	4	2	**CB**	.	.	.	2647.96
63	4	3	**CC**	.	.	.	2996.84
64	4	4	**CD**	.	.	.	4498.20
65	4	5	**CE**	.	.	.	3953.32
66	4	6	**D1**	.	.	.	3722.04
67	4	7	**DB**	.	.	.	3004.68
68	4	8	**YA**	.	.	.	3320.24
69	4	9	**DC**	.	.	.	3318.28
70	4	10	**YB**	.	.	.	3898.44
71	4	11	**YC**	.	.	.	3382.96
72	4	12	**DD**	.	.	.	3967.04
73	4	13	**DE**	.	.	.	5156.76
74	4	14	**DF**	.	.	.	4086.60
75	4	15	**DG**	.	.	.	4747.12
76	4	16	**DH**	.	.	.	2994.88
77	4	17	**Y1**	.	.	.	3782.80
78	4	18	**Y2**	.	.	.	3449.60
79	4	19	**YD**	.	.	.	3594.64
80	4	20	**Y3**	.	.	.	4121.88

* Codified due to confidentiality.

Table A4.4.3. Vredenburg 2006 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	91	125	72	3036.04
2	1	2	**CB**	96	135	68	3079.16
3	1	3	**CC**	90	125	68	3153.64
4	1	4	**CD**	76	120	72	3200.68
5	1	5	**CE**	89	140	70	2647.96
6	1	6	**D1**	90	125	70	2808.68
7	1	7	**DB**	91	140	70	2730.28
8	1	8	**YA**	92	125	74	2985.08
9	1	9	**DC**	90	120	70	2983.12
10	1	10	**YB**	86	120	74	4760.84
11	1	11	**YC**	95	120	72	4153.24
12	1	12	**DD**	90	130	74	4233.60
13	1	13	**DE**	89	110	68	3471.16
14	1	14	**DF**	84	130	70	3684.80
15	1	15	**DG**	92	125	70	3857.28
16	1	16	**DH**	88	125	68	3426.08
17	1	17	**Y1**	90	130	70	3418.24
18	1	18	**Y2**	93	130	72	1995.28
19	1	19	**YD**	90	130	70	1826.72
20	1	20	**Y3**	91	125	68	2432.36
21	2	1	**CA**	.	.	.	2197.16
22	2	2	**CB**	.	.	.	3204.60
23	2	3	**CC**	.	.	.	4049.36
24	2	4	**CD**	.	.	.	4756.92
25	2	5	**CE**	.	.	.	3220.28
26	2	6	**D1**	.	.	.	3806.32
27	2	7	**DB**	.	.	.	3114.44
28	2	8	**YA**	.	.	.	3841.60
29	2	9	**DC**	.	.	.	3422.16
30	2	10	**YB**	.	.	.	4155.20
31	2	11	**YC**	.	.	.	2702.84
32	2	12	**DD**	.	.	.	3326.12
33	2	13	**DE**	.	.	.	3171.28
34	2	14	**DF**	.	.	.	3281.04
35	2	15	**DG**	.	.	.	3365.32
36	2	16	**DH**	.	.	.	3537.80
37	2	17	**Y1**	.	.	.	2142.28
38	2	18	**Y2**	.	.	.	3398.64
39	2	19	**YD**	.	.	.	3192.84
40	2	20	**Y3**	.	.	.	3296.72

Table A4.4.3. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	.	3994.48
42	3	2	**CB**	.	.	.	3053.68
43	3	3	**CC**	.	.	.	3253.60
44	3	4	**CD**	.	.	.	3473.12
45	3	5	**CE**	.	.	.	3531.92
46	3	6	**D1**	.	.	.	3216.36
47	3	7	**DB**	.	.	.	1799.28
48	3	8	**YA**	.	.	.	2367.68
49	3	9	**DC**	.	.	.	2820.44
50	3	10	**YB**	.	.	.	4511.92
51	3	11	**YC**	.	.	.	5213.60
52	3	12	**DD**	.	.	.	3463.32
53	3	13	**DE**	.	.	.	3620.12
54	3	14	**DF**	.	.	.	3188.92
55	3	15	**DG**	.	.	.	2894.92
56	3	16	**DH**	.	.	.	2779.28
57	3	17	**Y1**	.	.	.	1685.60
58	3	18	**Y2**	.	.	.	2875.32
59	3	19	**YD**	.	.	.	3365.32
60	3	20	**Y3**	.	.	.	3112.48
61	4	1	**CA**	.	.	.	3431.96
62	4	2	**CB**	.	.	.	2608.76
63	4	3	**CC**	.	.	.	2103.08
64	4	4	**CD**	.	.	.	3283.00
65	4	5	**CE**	.	.	.	3326.12
66	4	6	**D1**	.	.	.	2393.16
67	4	7	**DB**	.	.	.	3041.92
68	4	8	**YA**	.	.	.	3563.28
69	4	9	**DC**	.	.	.	3814.16
70	4	10	**YB**	.	.	.	3655.40
71	4	11	**YC**	.	.	.	4337.48
72	4	12	**DD**	.	.	.	3747.52
73	4	13	**DE**	.	.	.	2303.00
74	4	14	**DF**	.	.	.	3106.60
75	4	15	**DG**	.	.	.	3075.24
76	4	16	**DH**	.	.	.	1618.96
77	4	17	**Y1**	.	.	.	1969.80
78	4	18	**Y2**	.	.	.	3304.56
79	4	19	**YD**	.	.	.	1830.64
80	4	20	**Y3**	.	.	.	2130.52

* Codified due to confidentiality.

Table A4.4.4. Roodebloem 2006 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	106	115	72	6128.92
2	1	2	**CB**	111	135	70	3945.48
3	1	3	**CC**	106	125	68	4429.60
4	1	4	**CD**	89	120	72	4778.48
5	1	5	**CE**	104	140	70	4864.72
6	1	6	**D1**	106	125	70	4611.88
7	1	7	**DB**	108	150	70	5239.08
8	1	8	**YA**	107	125	76	4666.76
9	1	9	**DC**	106	115	68	3957.24
10	1	10	**YB**	106	120	74	4441.36
11	1	11	**YC**	110	120	72	5419.40
12	1	12	**DD**	106	140	72	3959.20
13	1	13	**DE**	105	115	68	5190.08
14	1	14	**DF**	103	140	72	6050.52
15	1	15	**DG**	112	135	72	5513.48
16	1	16	**DH**	105	130	72	5605.60
17	1	17	**Y1**	105	125	72	6232.80
18	1	18	**Y2**	108	130	74	5503.68
19	1	19	**YD**	105	125	70	5484.08
20	1	20	**Y3**	106	125	74	6195.56
21	2	1	**CA**	.	.	.	6085.80
22	2	2	**CB**	.	.	.	4807.88
23	2	3	**CC**	.	.	.	5486.04
24	2	4	**CD**	.	.	.	5309.64
25	2	5	**CE**	.	.	.	5497.80
26	2	6	**D1**	.	.	.	4992.12
27	2	7	**DB**	.	.	.	5913.32
28	2	8	**YA**	.	.	.	6293.56
29	2	9	**DC**	.	.	.	4468.80
30	2	10	**YB**	.	.	.	7144.20
31	2	11	**YC**	.	.	.	6183.80
32	2	12	**DD**	.	.	.	4852.96
33	2	13	**DE**	.	.	.	6058.36
34	2	14	**DF**	.	.	.	4937.24
35	2	15	**DG**	.	.	.	5689.88
36	2	16	**DH**	.	.	.	6201.44
37	2	17	**Y1**	.	.	.	5538.96
38	2	18	**Y2**	.	.	.	5760.44
39	2	19	**YD**	.	.	.	5468.40
40	2	20	**Y3**	.	.	.	5731.04

Table A4.4.4. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	.	5899.60
42	3	2	**CB**	.	.	.	5260.64
43	3	3	**CC**	.	.	.	4613.84
44	3	4	**CD**	.	.	.	5323.36
45	3	5	**CE**	.	.	.	5829.04
46	3	6	**D1**	.	.	.	5327.28
47	3	7	**DB**	.	.	.	5966.24
48	3	8	**YA**	.	.	.	6287.68
49	3	9	**DC**	.	.	.	5497.80
50	3	10	**YB**	.	.	.	6609.12
51	3	11	**YC**	.	.	.	5842.76
52	3	12	**DD**	.	.	.	5468.40
53	3	13	**DE**	.	.	.	5096.00
54	3	14	**DF**	.	.	.	5717.32
55	3	15	**DG**	.	.	.	6111.28
56	3	16	**DH**	.	.	.	5580.12
57	3	17	**Y1**	.	.	.	5895.68
58	3	18	**Y2**	.	.	.	5799.64
59	3	19	**YD**	.	.	.	5689.88
60	3	20	**Y3**	.	.	.	4717.72
61	4	1	**CA**	.	.	.	6728.68
62	4	2	**CB**	.	.	.	5599.72
63	4	3	**CC**	.	.	.	5711.44
64	4	4	**CD**	.	.	.	5880.00
65	4	5	**CE**	.	.	.	5760.44
66	4	6	**D1**	.	.	.	5058.76
67	4	7	**DB**	.	.	.	6138.72
68	4	8	**YA**	.	.	.	6336.68
69	4	9	**DC**	.	.	.	5252.80
70	4	10	**YB**	.	.	.	6567.96
71	4	11	**YC**	.	.	.	6360.20
72	4	12	**DD**	.	.	.	6307.28
73	4	13	**DE**	.	.	.	5970.16
74	4	14	**DF**	.	.	.	4833.36
75	4	15	**DG**	.	.	.	4315.92
76	4	16	**DH**	.	.	.	6430.76
77	4	17	**Y1**	.	.	.	6469.96
78	4	18	**Y2**	.	.	.	5762.40
79	4	19	**YD**	.	.	.	5750.64
80	4	20	**Y3**	.	.	.	6679.68

* Codified due to confidentiality.

Table A4.4.5. Tygerhoek 2006 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	102	105	66	4707.92
2	1	2	**CB**	112	125	64	5092.08
3	1	3	**CC**	110	115	64	4915.68
4	1	4	**CD**	86	105	66	3388.84
5	1	5	**CE**	99	135	66	4233.60
6	1	6	**D1**	101	115	64	3737.72
7	1	7	**DB**	103	125	64	4380.60
8	1	8	**YA**	98	120	72	4153.24
9	1	9	**DC**	102	110	64	3876.88
10	1	10	**YB**	98	110	68	4690.28
11	1	11	**YC**	104	120	70	5003.88
12	1	12	**DD**	101	120	70	3800.44
13	1	13	**DE**	102	110	64	4155.20
14	1	14	**DF**	95	130	70	4480.56
15	1	15	**DG**	107	125	68	4535.44
16	1	16	**DH**	102	125	66	4235.56
17	1	17	**Y1**	99	115	68	4509.96
18	1	18	**Y2**	101	120	72	4219.88
19	1	19	**YD**	99	110	68	4464.88
20	1	20	**Y3**	101	115	70	4464.88
21	2	1	**CA**	.	.	.	4472.72
22	2	2	**CB**	.	.	.	3665.20
23	2	3	**CC**	.	.	.	4280.64
24	2	4	**CD**	.	.	.	2812.60
25	2	5	**CE**	.	.	.	3814.16
26	2	6	**D1**	.	.	.	3457.44
27	2	7	**DB**	.	.	.	4151.28
28	2	8	**YA**	.	.	.	4188.52
29	2	9	**DC**	.	.	.	3990.56
30	2	10	**YB**	.	.	.	4323.76
31	2	11	**YC**	.	.	.	4592.28
32	2	12	**DD**	.	.	.	3910.20
33	2	13	**DE**	.	.	.	4180.68
34	2	14	**DF**	.	.	.	3931.76
35	2	15	**DG**	.	.	.	3363.36
36	2	16	**DH**	.	.	.	3943.52
37	2	17	**Y1**	.	.	.	3833.76
38	2	18	**Y2**	.	.	.	4282.60
39	2	19	**YD**	.	.	.	4082.68
40	2	20	**Y3**	.	.	.	3933.72

Table A4.4.5. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	.	3798.48
42	3	2	**CB**	.	.	.	4296.32
43	3	3	**CC**	.	.	.	4082.68
44	3	4	**CD**	.	.	.	2491.16
45	3	5	**CE**	.	.	.	3980.76
46	3	6	**D1**	.	.	.	3651.48
47	3	7	**DB**	.	.	.	2802.80
48	3	8	**YA**	.	.	.	4188.52
49	3	9	**DC**	.	.	.	4176.76
50	3	10	**YB**	.	.	.	4798.08
51	3	11	**YC**	.	.	.	5082.28
52	3	12	**DD**	.	.	.	3645.60
53	3	13	**DE**	.	.	.	3767.12
54	3	14	**DF**	.	.	.	4117.96
55	3	15	**DG**	.	.	.	3451.56
56	3	16	**DH**	.	.	.	3549.56
57	3	17	**Y1**	.	.	.	4061.12
58	3	18	**Y2**	.	.	.	4494.28
59	3	19	**YD**	.	.	.	3857.28
60	3	20	**Y3**	.	.	.	3967.04
61	4	1	**CA**	.	.	.	3696.56
62	4	2	**CB**	.	.	.	3627.96
63	4	3	**CC**	.	.	.	4200.28
64	4	4	**CD**	.	.	.	3145.80
65	4	5	**CE**	.	.	.	3567.20
66	4	6	**D1**	.	.	.	3484.88
67	4	7	**DB**	.	.	.	4108.16
68	4	8	**YA**	.	.	.	4400.20
69	4	9	**DC**	.	.	.	4384.52
70	4	10	**YB**	.	.	.	5217.52
71	4	11	**YC**	.	.	.	4815.72
72	4	12	**DD**	.	.	.	3320.24
73	4	13	**DE**	.	.	.	5072.48
74	4	14	**DF**	.	.	.	3441.76
75	4	15	**DG**	.	.	.	3696.56
76	4	16	**DH**	.	.	.	3332.00
77	4	17	**Y1**	.	.	.	4484.48
78	4	18	**Y2**	.	.	.	5658.52
79	4	19	**YD**	.	.	.	4519.76
80	4	20	**Y3**	.	.	.	4558.96

* Codified due to confidentiality.

Table A4.4.6. Napier 2006 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	113	115	72	4764.76
2	1	2	**CB**	118	125	72	3925.88
3	1	3	**CC**	111	120	70	5225.36
4	1	4	**CD**	89	105	68	3892.56
5	1	5	**CE**	108	130	72	4019.96
6	1	6	**D1**	113	105	72	5227.32
7	1	7	**DB**	114	125	72	5707.52
8	1	8	**YA**	109	110	76	4845.12
9	1	9	**DC**	107	100	70	4711.84
10	1	10	**YB**	107	110	74	4825.52
11	1	11	**YC**	114	115	74	5025.44
12	1	12	**DD**	113	110	74	4982.32
13	1	13	**DE**	106	100	70	4625.60
14	1	14	**DF**	101	110	72	4159.12
15	1	15	**DG**	119	120	70	4349.24
16	1	16	**DH**	107	115	72	5121.48
17	1	17	**Y1**	107	115	70	5037.20
18	1	18	**Y2**	110	115	74	5948.60
19	1	19	**YD**	108	110	74	4737.32
20	1	20	**Y3**	109	110	74	4784.36
21	2	1	**CA**	.	.	.	5268.48
22	2	2	**CB**	.	.	.	4668.72
23	2	3	**CC**	.	.	.	6340.60
24	2	4	**CD**	.	.	.	4339.44
25	2	5	**CE**	.	.	.	4937.24
26	2	6	**D1**	.	.	.	6536.60
27	2	7	**DB**	.	.	.	5305.72
28	2	8	**YA**	.	.	.	5497.80
29	2	9	**DC**	.	.	.	5946.64
30	2	10	**YB**	.	.	.	2346.12
31	2	11	**YC**	.	.	.	5742.80
32	2	12	**DD**	.	.	.	5339.04
33	2	13	**DE**	.	.	.	5970.16
34	2	14	**DF**	.	.	.	4988.20
35	2	15	**DG**	.	.	.	3557.40
36	2	16	**DH**	.	.	.	5143.04
37	2	17	**Y1**	.	.	.	6152.44
38	2	18	**Y2**	.	.	.	5974.08
39	2	19	**YD**	.	.	.	4335.52
40	2	20	**Y3**	.	.	.	6270.04

Table A4.4.6. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	.	5552.68
42	3	2	**CB**	.	.	.	4594.24
43	3	3	**CC**	.	.	.	5515.44
44	3	4	**CD**	.	.	.	3382.96
45	3	5	**CE**	.	.	.	5468.40
46	3	6	**D1**	.	.	.	5684.00
47	3	7	**DB**	.	.	.	4676.56
48	3	8	**YA**	.	.	.	5517.40
49	3	9	**DC**	.	.	.	5493.88
50	3	10	**YB**	.	.	.	4531.52
51	3	11	**YC**	.	.	.	5729.08
52	3	12	**DD**	.	.	.	4480.56
53	3	13	**DE**	.	.	.	5466.44
54	3	14	**DF**	.	.	.	4903.92
55	3	15	**DG**	.	.	.	4596.20
56	3	16	**DH**	.	.	.	5760.44
57	3	17	**Y1**	.	.	.	5762.40
58	3	18	**Y2**	.	.	.	5693.80
59	3	19	**YD**	.	.	.	5664.40
60	3	20	**Y3**	.	.	.	6730.64
61	4	1	**CA**	.	.	.	5464.48
62	4	2	**CB**	.	.	.	4760.84
63	4	3	**CC**	.	.	.	5727.12
64	4	4	**CD**	.	.	.	4016.04
65	4	5	**CE**	.	.	.	4792.20
66	4	6	**D1**	.	.	.	5442.92
67	4	7	**DB**	.	.	.	4896.08
68	4	8	**YA**	.	.	.	5944.68
69	4	9	**DC**	.	.	.	5092.08
70	4	10	**YB**	.	.	.	3692.64
71	4	11	**YC**	.	.	.	6552.28
72	4	12	**DD**	.	.	.	5223.40
73	4	13	**DE**	.	.	.	4772.60
74	4	14	**DF**	.	.	.	4484.48
75	4	15	**DG**	.	.	.	3794.56
76	4	16	**DH**	.	.	.	5225.36
77	4	17	**Y1**	.	.	.	6101.48
78	4	18	**Y2**	.	.	.	6934.48
79	4	19	**YD**	.	.	.	6015.24
80	4	20	**Y3**	.	.	.	6548.36

* Codified due to confidentiality.

Table A4.4.7. Mariendahl 2007 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	100	.	74	3204.60
2	1	2	**CC**	99	.	70	2391.20
3	1	3	**CE**	100	.	72	3088.96
4	1	4	**CD**	87	.	70	1515.08
5	1	5	**D1**	97	.	70	4472.72
6	1	6	**D2**	96	.	70	5429.20
7	1	7	**D3**	97	.	70	3067.40
8	1	8	**D4**	97	.	70	2685.20
9	1	9	**YC**	103	.	70	2250.08
10	1	10	**Y2**	97	.	76	3488.80
11	1	11	**EA**	97	.	76	3357.48
12	1	12	**EB**	97	.	74	2638.16
13	1	13	**G1**	95	.	72	3106.60
14	1	14	**G2**	102	.	72	3426.08
15	1	15	**H1**	100	.	72	4557.00
16	1	16	**H2**	98	.	70	3773.00
17	1	17	**H3**	102	.	72	3637.76
18	1	18	**H4**	97	.	72	2561.72
19	1	19	**H5**	99	.	72	2303.00
20	1	20	**H6**	97	.	72	3449.60
21	2	1	**CA**	.	.	72	2708.72
22	2	2	**CC**	.	.	72	3908.24
23	2	3	**CE**	.	.	70	3600.52
24	2	4	**CD**	.	.	70	1742.44
25	2	5	**D1**	.	.	70	6573.84
26	2	6	**D2**	.	.	70	3618.16
27	2	7	**D3**	.	.	72	3208.52
28	2	8	**D4**	.	.	70	3528.00
29	2	9	**YC**	.	.	72	2581.32
30	2	10	**Y2**	.	.	74	4186.56
31	2	11	**EA**	.	.	74	6050.52
32	2	12	**EB**	.	.	76	2840.04
33	2	13	**G1**	.	.	72	2657.76
34	2	14	**G2**	.	.	70	3373.16
35	2	15	**H1**	.	.	72	3381.00
36	2	16	**H2**	.	.	70	3116.40
37	2	17	**H3**	.	.	72	3361.40
38	2	18	**H4**	.	.	72	2724.40
39	2	19	**H5**	.	.	72	3337.88
40	2	20	**H6**	.	.	72	3700.48

Table A4.4.7. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	74	3083.08
42	3	2	**CC**	.	.	70	3949.40
43	3	3	**CE**	.	.	72	3426.08
44	3	4	**CD**	.	.	68	3065.44
45	3	5	**D1**	.	.	70	4619.72
46	3	6	**D2**	.	.	70	3737.72
47	3	7	**D3**	.	.	68	4253.20
48	3	8	**D4**	.	.	70	4176.76
49	3	9	**YC**	.	.	68	3535.84
50	3	10	**Y2**	.	.	76	4102.28
51	3	11	**EA**	.	.	74	4168.92
52	3	12	**EB**	.	.	74	2808.68
53	3	13	**G1**	.	.	72	3388.84
54	3	14	**G2**	.	.	72	4027.80
55	3	15	**H1**	.	.	70	3345.72
56	3	16	**H2**	.	.	70	4894.12
57	3	17	**H3**	.	.	72	5325.32
58	3	18	**H4**	.	.	72	3373.16
59	3	19	**H5**	.	.	72	3843.56
60	3	20	**H6**	.	.	70	3036.04

* Codified due to confidentiality.

Table A4.4.8. Langgewens 2007 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	91	100	74	5744.76
2	1	2	**CC**	91	112	70	5254.76
3	1	3	**CE**	91	114	70	5772.20
4	1	4	**CD**	76	95	74	4725.56
5	1	5	**D1**	87	120	72	6268.08
6	1	6	**D2**	87	118	72	6317.08
7	1	7	**D3**	91	110	74	4854.92
8	1	8	**D4**	91	103	72	5115.60
9	1	9	**YC**	93	94	74	5970.16
10	1	10	**Y2**	87	112	76	6430.76
11	1	11	**EA**	97	125	74	7155.96
12	1	12	**EB**	85	128	76	5137.16
13	1	13	**G1**	91	107	74	7275.52
14	1	14	**G2**	89	97	74	6591.48
15	1	15	**H1**	91	128	74	6893.32
16	1	16	**H2**	87	127	72	5831.00
17	1	17	**H3**	91	115	74	5670.28
18	1	18	**H4**	88	118	74	4956.84
19	1	19	**H5**	89	120	72	4094.44
20	1	20	**H6**	88	113	74	4917.64
21	2	1	**CA**	.	.	74	5901.56
22	2	2	**CC**	.	.	70	5591.88
23	2	3	**CE**	.	.	72	4239.48
24	2	4	**CD**	.	.	76	4423.72
25	2	5	**D1**	.	.	72	5948.60
26	2	6	**D2**	.	.	74	5946.64
27	2	7	**D3**	.	.	74	5246.92
28	2	8	**D4**	.	.	72	5007.80
29	2	9	**YC**	.	.	74	5793.76
30	2	10	**Y2**	.	.	76	4668.72
31	2	11	**EA**	.	.	74	5431.16
32	2	12	**EB**	.	.	76	5956.44
33	2	13	**G1**	.	.	74	4994.08
34	2	14	**G2**	.	.	74	5899.60
35	2	15	**H1**	.	.	74	5891.76
36	2	16	**H2**	.	.	74	5293.96
37	2	17	**H3**	.	.	74	6503.28
38	2	18	**H4**	.	.	74	5756.52
39	2	19	**H5**	.	.	72	5785.92
40	2	20	**H6**	.	.	74	4770.64

Table A4.4.8. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	74	6589.52
42	3	2	**CC**	.	.	72	6091.68
43	3	3	**CE**	.	.	72	4872.56
44	3	4	**CD**	.	.	74	4864.72
45	3	5	**D1**	.	.	72	6807.08
46	3	6	**D2**	.	.	72	7230.44
47	3	7	**D3**	.	.	74	6783.56
48	3	8	**D4**	.	.	72	6244.56
49	3	9	**YC**	.	.	74	4570.72
50	3	10	**Y2**	.	.	76	6534.64
51	3	11	**EA**	.	.	74	6405.28
52	3	12	**EB**	.	.	76	6093.64
53	3	13	**G1**	.	.	74	6722.80
54	3	14	**G2**	.	.	74	6138.72
55	3	15	**H1**	.	.	74	5378.24
56	3	16	**H2**	.	.	72	6399.40
57	3	17	**H3**	.	.	74	5364.52
58	3	18	**H4**	.	.	74	5938.80
59	3	19	**H5**	.	.	72	4704.00
60	3	20	**H6**	.	.	74	6105.40

* Codified due to confidentiality.

Table A4.4.9. Klipheuwel 2007 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	92	110	70	6005.44
2	1	2	**CC**	95	116	68	6370.00
3	1	3	**CE**	89	130	68	5883.92
4	1	4	**CD**	78	97	74	5848.64
5	1	5	**D1**	93	115	70	5605.60
6	1	6	**D2**	93	115	70	6734.56
7	1	7	**D3**	95	120	72	5989.76
8	1	8	**D4**	95	118	70	5670.28
9	1	9	**YC**	97	110	72	4923.52
10	1	10	**Y2**	95	121	72	6036.80
11	1	11	**EA**	89	130	72	6152.44
12	1	12	**EB**	88	130	72	4809.84
13	1	13	**G1**	78	107	74	4984.28
14	1	14	**G2**	87	110	72	6622.84
15	1	15	**H1**	89	132	72	6191.64
16	1	16	**H2**	89	130	70	5640.88
17	1	17	**H3**	89	128	72	4598.16
18	1	18	**H4**	90	122	72	5431.16
19	1	19	**H5**	87	133	72	5597.76
20	1	20	**H6**	85	118	74	4555.04
21	2	1	**CA**	.	.	70	5272.40
22	2	2	**CC**	.	.	68	3298.68
23	2	3	**CE**	.	.	70	3547.60
24	2	4	**CD**	.	.	74	5546.80
25	2	5	**D1**	.	.	70	4327.68
26	2	6	**D2**	.	.	70	4021.92
27	2	7	**D3**	.	.	74	4968.60
28	2	8	**D4**	.	.	70	4057.20
29	2	9	**YC**	.	.	72	6707.12
30	2	10	**Y2**	.	.	72	3096.80
31	2	11	**EA**	.	.	72	4174.80
32	2	12	**EB**	.	.	72	5399.80
33	2	13	**G1**	.	.	74	4263.00
34	2	14	**G2**	.	.	72	3286.92
35	2	15	**H1**	.	.	70	4010.16
36	2	16	**H2**	.	.	70	3288.88
37	2	17	**H3**	.	.	72	4374.72
38	2	18	**H4**	.	.	72	4888.24
39	2	19	**H5**	.	.	72	4482.52
40	2	20	**H6**	.	.	72	3918.04

Table A4.4.9. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	72	4231.64
42	3	2	**CC**	.	.	70	3814.16
43	3	3	**CE**	.	.	70	3680.88
44	3	4	**CD**	.	.	76	5725.16
45	3	5	**D1**	.	.	72	3653.44
46	3	6	**D2**	.	.	72	5593.84
47	3	7	**D3**	.	.	72	3359.44
48	3	8	**D4**	.	.	70	3720.08
49	3	9	**YC**	.	.	72	2459.80
50	3	10	**Y2**	.	.	74	5309.64
51	3	11	**EA**	.	.	72	4298.28
52	3	12	**EB**	.	.	74	3557.40
53	3	13	**G1**	.	.	74	3004.68
54	3	14	**G2**	.	.	72	2775.36
55	3	15	**H1**	.	.	70	2981.16
56	3	16	**H2**	.	.	70	4490.36
57	3	17	**H3**	.	.	72	4511.92
58	3	18	**H4**	.	.	74	5495.84
59	3	19	**H5**	.	.	72	4227.72
60	3	20	**H6**	.	.	72	2696.96

* Codified due to confidentiality.

Table A4.4.10. Piketberg 2007 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	104	110	76	4325.72
2	1	2	**CC**	103	105	74	5121.48
3	1	3	**CE**	101	135	74	5017.60
4	1	4	**CD**	90	95	78	4378.64
5	1	5	**D1**	97	115	74	4662.84
6	1	6	**D2**	98	110	72	5092.08
7	1	7	**D3**	99	120	74	5625.20
8	1	8	**D4**	100	120	72	4241.44
9	1	9	**YC**	103	120	76	4909.80
10	1	10	**Y2**	98	125	76	5203.80
11	1	11	**EA**	98	140	76	5062.68
12	1	12	**EB**	98	125	78	3467.24
13	1	13	**G1**	95	90	76	3524.08
14	1	14	**G2**	102	105	74	5050.92
15	1	15	**H1**	99	140	70	4760.84
16	1	16	**H2**	99	145	74	4631.48
17	1	17	**H3**	101	145	78	4178.72
18	1	18	**H4**	98	130	78	4656.96
19	1	19	**H5**	99	140	76	4476.64
20	1	20	**H6**	100	105	72	4241.44
21	2	1	**CA**	.	.	76	6023.08
22	2	2	**CC**	.	.	74	4186.56
23	2	3	**CE**	.	.	74	4713.80
24	2	4	**CD**	.	.	78	5195.96
25	2	5	**D1**	.	.	74	5225.36
26	2	6	**D2**	.	.	72	4986.24
27	2	7	**D3**	.	.	74	4760.84
28	2	8	**D4**	.	.	72	4455.08
29	2	9	**YC**	.	.	76	4911.76
30	2	10	**Y2**	.	.	78	4560.92
31	2	11	**EA**	.	.	76	3627.96
32	2	12	**EB**	.	.	78	3863.16
33	2	13	**G1**	.	.	76	5284.16
34	2	14	**G2**	.	.	76	5856.48
35	2	15	**H1**	.	.	74	5005.84
36	2	16	**H2**	.	.	74	5499.76
37	2	17	**H3**	.	.	76	5119.52
38	2	18	**H4**	.	.	76	4557.00
39	2	19	**H5**	.	.	76	4545.24
40	2	20	**H6**	.	.	74	4317.88

Table A4.4.10. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	76	6656.16
42	3	2	**CC**	.	.	74	4315.92
43	3	3	**CE**	.	.	74	4506.04
44	3	4	**CD**	.	.	78	6477.80
45	3	5	**D1**	.	.	74	4760.84
46	3	6	**D2**	.	.	74	5595.80
47	3	7	**D3**	.	.	74	6818.84
48	3	8	**D4**	.	.	72	4872.56
49	3	9	**YC**	.	.	76	5895.68
50	3	10	**Y2**	.	.	78	5323.36
51	3	11	**EA**	.	.	76	4760.84
52	3	12	**EB**	.	.	78	4100.32
53	3	13	**G1**	.	.	76	5321.40
54	3	14	**G2**	.	.	76	5803.56
55	3	15	**H1**	.	.	76	5993.68
56	3	16	**H2**	.	.	74	4831.40
57	3	17	**H3**	.	.	76	5260.64
58	3	18	**H4**	.	.	76	6667.92
59	3	19	**H5**	.	.	76	5170.48
60	3	20	**H6**	.	.	74	4984.28

* Codified due to confidentiality.

Table A4.4.11. Roodebloem 2007 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	103	130	76	7193.20
2	1	2	**CC**	105	126	72	6062.28
3	1	3	**CE**	105	140	72	5356.68
4	1	4	**CD**	90	130	76	5927.04
5	1	5	**D1**	105	125	74	7163.80
6	1	6	**D2**	104	120	72	6777.68
7	1	7	**D3**	104	125	68	7281.40
8	1	8	**D4**	104	123	70	7322.56
9	1	9	**YC**	109	116	66	6528.76
10	1	10	**Y2**	103	121	76	5962.32
11	1	11	**EA**	105	131	74	5511.52
12	1	12	**EB**	104	131	76	5331.20
13	1	13	**G1**	100	126	76	8147.72
14	1	14	**G2**	107	120	78	7069.72
15	1	15	**H1**	105	140	74	6234.76
16	1	16	**H2**	102	140	74	7048.16
17	1	17	**H3**	107	140	74	7861.56
18	1	18	**H4**	103	136	76	5319.44
19	1	19	**H5**	104	148	74	7771.40
20	1	20	**H6**	104	129	74	7840.00
21	2	1	**CA**	.	.	76	7512.68
22	2	2	**CC**	.	.	72	7406.84
23	2	3	**CE**	.	.	72	6054.44
24	2	4	**CD**	.	.	76	6483.68
25	2	5	**D1**	.	.	72	7267.68
26	2	6	**D2**	.	.	74	6628.72
27	2	7	**D3**	.	.	74	7557.76
28	2	8	**D4**	.	.	72	7269.64
29	2	9	**YC**	.	.	72	7736.12
30	2	10	**Y2**	.	.	76	5017.60
31	2	11	**EA**	.	.	74	5874.12
32	2	12	**EB**	.	.	76	5674.20
33	2	13	**G1**	.	.	74	7575.40
34	2	14	**G2**	.	.	76	8263.36
35	2	15	**H1**	.	.	74	6299.44
36	2	16	**H2**	.	.	74	6005.44
37	2	17	**H3**	.	.	74	4958.80
38	2	18	**H4**	.	.	76	5997.60
39	2	19	**H5**	.	.	74	6268.08
40	2	20	**H6**	.	.	72	6632.64

Table A4.4.11. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	76	6903.12
42	3	2	**CC**	.	.	72	7230.44
43	3	3	**CE**	.	.	72	6011.32
44	3	4	**CD**	.	.	78	6336.68
45	3	5	**D1**	.	.	74	5905.48
46	3	6	**D2**	.	.	74	7510.72
47	3	7	**D3**	.	.	74	6832.56
48	3	8	**D4**	.	.	72	7408.80
49	3	9	**YC**	.	.	74	7622.44
50	3	10	**Y2**	.	.	76	6170.08
51	3	11	**EA**	.	.	76	6027.00
52	3	12	**EB**	.	.	76	5056.80
53	3	13	**G1**	.	.	76	6632.64
54	3	14	**G2**	.	.	76	9066.96
55	3	15	**H1**	.	.	74	4968.60
56	3	16	**H2**	.	.	74	6485.64
57	3	17	**H3**	.	.	74	6277.88
58	3	18	**H4**	.	.	76	6203.40
59	3	19	**H5**	.	.	74	4166.96
60	3	20	**H6**	.	.	72	6401.36

* Codified due to confidentiality.

Table A4.4.12. Tygerhoek 2007 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	104	115	74	6397.44
2	1	2	**CC**	106	121	74	5783.96
3	1	3	**CE**	105	132	74	5566.40
4	1	4	**CD**	87	122	72	3780.84
5	1	5	**D1**	102	136	74	7400.96
6	1	6	**D2**	106	126	74	5787.88
7	1	7	**D3**	105	122	76	5505.64
8	1	8	**D4**	103	122	74	6154.40
9	1	9	**YC**	106	119	74	6885.48
10	1	10	**Y2**	106	119	76	5997.60
11	1	11	**EA**	107	135	76	6483.68
12	1	12	**EB**	105	135	78	5401.76
13	1	13	**G1**	102	114	76	5305.72
14	1	14	**G2**	103	110	74	5327.28
15	1	15	**H1**	104	133	74	4545.24
16	1	16	**H2**	105	135	74	5811.40
17	1	17	**H3**	105	138	76	6503.28
18	1	18	**H4**	102	128	76	5617.36
19	1	19	**H5**	107	134	74	2606.80
20	1	20	**H6**	102	112	76	6068.16
21	2	1	**CA**	.	.	76	6142.64
22	2	2	**CC**	.	.	72	5862.36
23	2	3	**CE**	.	.	74	5335.12
24	2	4	**CD**	.	.	72	4476.64
25	2	5	**D1**	.	.	74	5819.24
26	2	6	**D2**	.	.	74	6483.68
27	2	7	**D3**	.	.	76	6714.96
28	2	8	**D4**	.	.	74	5468.40
29	2	9	**YC**	.	.	76	6530.72
30	2	10	**Y2**	.	.	78	6027.00
31	2	11	**EA**	.	.	76	6783.56
32	2	12	**EB**	.	.	78	5346.88
33	2	13	**G1**	.	.	78	5576.20
34	2	14	**G2**	.	.	76	7408.80
35	2	15	**H1**	.	.	76	5954.48
36	2	16	**H2**	.	.	76	5727.12
37	2	17	**H3**	.	.	76	6595.40
38	2	18	**H4**	.	.	76	6032.88
39	2	19	**H5**	.	.	76	5458.60
40	2	20	**H6**	.	.	78	6272.00

Table A4.4.12. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	74	4067.00
42	3	2	**CC**	.	.	72	5838.84
43	3	3	**CE**	.	.	74	5525.24
44	3	4	**CD**	.	.	70	3045.84
45	3	5	**D1**	.	.	74	5480.16
46	3	6	**D2**	.	.	74	5035.24
47	3	7	**D3**	.	.	74	5258.68
48	3	8	**D4**	.	.	74	5589.92
49	3	9	**YC**	.	.	76	6454.28
50	3	10	**Y2**	.	.	78	6505.24
51	3	11	**EA**	.	.	76	4999.96
52	3	12	**EB**	.	.	78	5368.44
53	3	13	**G1**	.	.	76	5495.84
54	3	14	**G2**	.	.	74	5664.40
55	3	15	**H1**	.	.	74	5993.68
56	3	16	**H2**	.	.	74	4598.16
57	3	17	**H3**	.	.	76	6191.64
58	3	18	**H4**	.	.	74	5215.56
59	3	19	**H5**	.	.	74	4621.68
60	3	20	**H6**	.	.	76	5987.80

* Codified due to confidentiality.

Table A4.4.13. Napier 2007 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	103	116	74	6750.24
2	1	2	**CC**	107	124	72	5842.76
3	1	3	**CE**	106	140	74	4529.56
4	1	4	**CD**	95	122	66	1326.92
5	1	5	**D1**	101	125	76	5552.68
6	1	6	**D2**	102	126	76	5664.40
7	1	7	**D3**	103	124	76	5407.64
8	1	8	**D4**	105	126	74	6722.80
9	1	9	**YC**	106	122	78	6083.84
10	1	10	**Y2**	101	128	78	6336.68
11	1	11	**EA**	102	132	76	5411.56
12	1	12	**EB**	101	133	78	5446.84
13	1	13	**G1**	99	127	76	5997.60
14	1	14	**G2**	105	114	76	5821.20
15	1	15	**H1**	103	141	76	6015.24
16	1	16	**H2**	101	151	74	7079.52
17	1	17	**H3**	104	151	76	6066.20
18	1	18	**H4**	101	137	76	3921.96
19	1	19	**H5**	102	130	74	3000.76
20	1	20	**H6**	101	126	76	7261.80
21	2	1	**CA**	.	.	72	5119.52
22	2	2	**CC**	.	.	72	6111.28
23	2	3	**CE**	.	.	74	5241.04
24	2	4	**CD**	.	.	.	284.20
25	2	5	**D1**	.	.	76	6656.16
26	2	6	**D2**	.	.	74	6540.52
27	2	7	**D3**	.	.	76	6197.52
28	2	8	**D4**	.	.	74	6224.96
29	2	9	**YC**	.	.	76	5697.72
30	2	10	**Y2**	.	.	78	6191.64
31	2	11	**EA**	.	.	76	5705.56
32	2	12	**EB**	.	.	78	4351.20
33	2	13	**G1**	.	.	76	6391.56
34	2	14	**G2**	.	.	74	6076.00
35	2	15	**H1**	.	.	76	6740.44
36	2	16	**H2**	.	.	76	6258.28
37	2	17	**H3**	.	.	76	5052.88
38	2	18	**H4**	.	.	76	6136.76
39	2	19	**H5**	.	.	76	4494.28
40	2	20	**H6**	.	.	76	6287.68

Table A4.4.13. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	74	6889.40
42	3	2	**CC**	.	.	72	5742.80
43	3	3	**CE**	.	.	74	5258.68
44	3	4	**CD**	.	.	.	427.28
45	3	5	**D1**	.	.	74	5437.04
46	3	6	**D2**	.	.	74	5962.32
47	3	7	**D3**	.	.	76	7146.16
48	3	8	**D4**	.	.	74	5793.76
49	3	9	**YC**	.	.	76	5007.80
50	3	10	**Y2**	.	.	78	6936.44
51	3	11	**EA**	.	.	76	5074.44
52	3	12	**EB**	.	.	78	3984.68
53	3	13	**G1**	.	.	74	5411.56
54	3	14	**G2**	.	.	76	6097.56
55	3	15	**H1**	.	.	76	6993.28
56	3	16	**H2**	.	.	74	6181.84
57	3	17	**H3**	.	.	76	6246.52
58	3	18	**H4**	.	.	76	4678.52
59	3	19	**H5**	.	.	74	4049.36
60	3	20	**H6**	.	.	76	6175.96

* Codified due to confidentiality.

Table A4.4.14. Riversdale 2007 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	104	130	72	3880.80
2	1	2	**CC**	106	143	68	4268.88
3	1	3	**CE**	106	169	72	4172.84
4	1	4	**CD**	92	150	70	2442.16
5	1	5	**D1**	103	123	70	3643.64
6	1	6	**D2**	104	143	72	5958.40
7	1	7	**D3**	106	156	72	5293.96
8	1	8	**D4**	106	138	70	6758.08
9	1	9	**YC**	109	142	74	7295.12
10	1	10	**Y2**	101	154	74	3982.72
11	1	11	**EA**	102	160	74	5501.72
12	1	12	**EB**	103	151	72	3128.16
13	1	13	**G1**	100	134	72	5021.52
14	1	14	**G2**	106	130	72	5807.48
15	1	15	**H1**	104	160	76	7010.92
16	1	16	**H2**	103	168	72	6840.40
17	1	17	**H3**	106	169	76	5721.24
18	1	18	**H4**	103	161	74	6079.92
19	1	19	**H5**	104	147	74	3925.88
20	1	20	**H6**	103	140	74	4464.88
21	2	1	**CA**	.	.	70	3920.00
22	2	2	**CC**	.	.	72	6389.60
23	2	3	**CE**	.	.	74	8069.32
24	2	4	**CD**	.	.	68	1611.12
25	2	5	**D1**	.	.	72	6558.16
26	2	6	**D2**	.	.	74	6348.44
27	2	7	**D3**	.	.	72	7232.40
28	2	8	**D4**	.	.	72	6360.20
29	2	9	**YC**	.	.	72	4198.32
30	2	10	**Y2**	.	.	76	4925.48
31	2	11	**EA**	.	.	74	4737.32
32	2	12	**EB**	.	.	76	5052.88
33	2	13	**G1**	.	.	74	5050.92
34	2	14	**G2**	.	.	72	5852.56
35	2	15	**H1**	.	.	72	4078.76
36	2	16	**H2**	.	.	74	5927.04
37	2	17	**H3**	.	.	74	6556.20
38	2	18	**H4**	.	.	74	5442.92
39	2	19	**H5**	.	.	74	4178.72
40	2	20	**H6**	.	.	76	6328.84

Table A4.4.14. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	68	3018.40
42	3	2	**CC**	.	.	70	5856.48
43	3	3	**CE**	.	.	70	6087.76
44	3	4	**CD**	.	.	.	1277.92
45	3	5	**D1**	.	.	72	5591.88
46	3	6	**D2**	.	.	72	6103.44
47	3	7	**D3**	.	.	72	4623.64
48	3	8	**D4**	.	.	72	5407.64
49	3	9	**YC**	.	.	72	6158.32
50	3	10	**Y2**	.	.	74	6066.20
51	3	11	**EA**	.	.	76	5501.72
52	3	12	**EB**	.	.	74	6226.92
53	3	13	**G1**	.	.	74	5476.24
54	3	14	**G2**	.	.	74	5850.60
55	3	15	**H1**	.	.	74	5638.92
56	3	16	**H2**	.	.	72	4635.40
57	3	17	**H3**	.	.	74	5080.32
58	3	18	**H4**	.	.	76	6113.24
59	3	19	**H5**	.	.	74	5425.28
60	3	20	**H6**	.	.	74	6221.04

* Codified due to confidentiality.

Table A4.4.15. Albertinia 2007 season elite breeding block trial

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
1	1	1	**CA**	103	113	70	5013.68
2	1	2	**CC**	104	124	70	6060.32
3	1	3	**CE**	105	145	70	5495.84
4	1	4	**CD**	93	103	68	2744.00
5	1	5	**D1**	101	123	70	6368.04
6	1	6	**D2**	102	117	72	5631.08
7	1	7	**D3**	106	128	74	6364.12
8	1	8	**D4**	107	117	72	6797.28
9	1	9	**YC**	108	106	72	5717.32
10	1	10	**Y2**	102	124	74	6760.04
11	1	11	**EA**	102	144	74	7083.44
12	1	12	**EB**	102	132	74	5578.16
13	1	13	**G1**	100	110	74	6432.72
14	1	14	**G2**	107	110	72	7808.64
15	1	15	**H1**	105	133	72	6340.60
16	1	16	**H2**	103	140	72	6658.12
17	1	17	**H3**	107	150	72	6219.08
18	1	18	**H4**	102	144	74	6493.48
19	1	19	**H5**	104	160	72	5309.64
20	1	20	**H6**	102	122	72	5927.04
21	2	1	**CA**	.	.	70	5750.64
22	2	2	**CC**	.	.	70	6665.96
23	2	3	**CE**	.	.	72	6295.52
24	2	4	**CD**	.	.	74	5399.80
25	2	5	**D1**	.	.	72	6595.40
26	2	6	**D2**	.	.	72	6573.84
27	2	7	**D3**	.	.	72	5460.56
28	2	8	**D4**	.	.	72	6483.68
29	2	9	**YC**	.	.	72	7105.00
30	2	10	**Y2**	.	.	74	6577.76
31	2	11	**EA**	.	.	74	6460.16
32	2	12	**EB**	.	.	76	5846.68
33	2	13	**G1**	.	.	74	7659.68
34	2	14	**G2**	.	.	74	6632.64
35	2	15	**H1**	.	.	74	7272.91
36	2	16	**H2**	.	.	74	6062.28
37	2	17	**H3**	.	.	74	6963.88
38	2	18	**H4**	.	.	74	5780.04
39	2	19	**H5**	.	.	72	5121.48
40	2	20	**H6**	.	.	74	5958.40

Table A4.4.15. (continued)

Plot	Block	Entry No.	Name*	Days to heading	Height, cm	Test weight, $kg.HL^{-1}$	Grain yield, $kg.ha^{-1}$
41	3	1	**CA**	.	.	70	5048.96
42	3	2	**CC**	.	.	70	7973.28
43	3	3	**CE**	.	.	72	5932.92
44	3	4	**CD**	.	.	74	5150.88
45	3	5	**D1**	.	.	72	6258.28
46	3	6	**D2**	.	.	72	7712.60
47	3	7	**D3**	.	.	74	6103.44
48	3	8	**D4**	.	.	70	6997.20
49	3	9	**YC**	.	.	74	7036.40
50	3	10	**Y2**	.	.	74	6348.44
51	3	11	**EA**	.	.	74	6789.44
52	3	12	**EB**	.	.	76	5891.76
53	3	13	**G1**	.	.	72	6401.36
54	3	14	**G2**	.	.	74	6638.52
55	3	15	**H1**	.	.	72	7295.12
56	3	16	**H2**	.	.	74	6650.28
57	3	17	**H3**	.	.	72	6381.76
58	3	18	**H4**	.	.	74	5478.20
59	3	19	**H5**	.	.	72	5672.24
60	3	20	**H6**	.	.	74	6054.44

* Codified due to confidentiality.

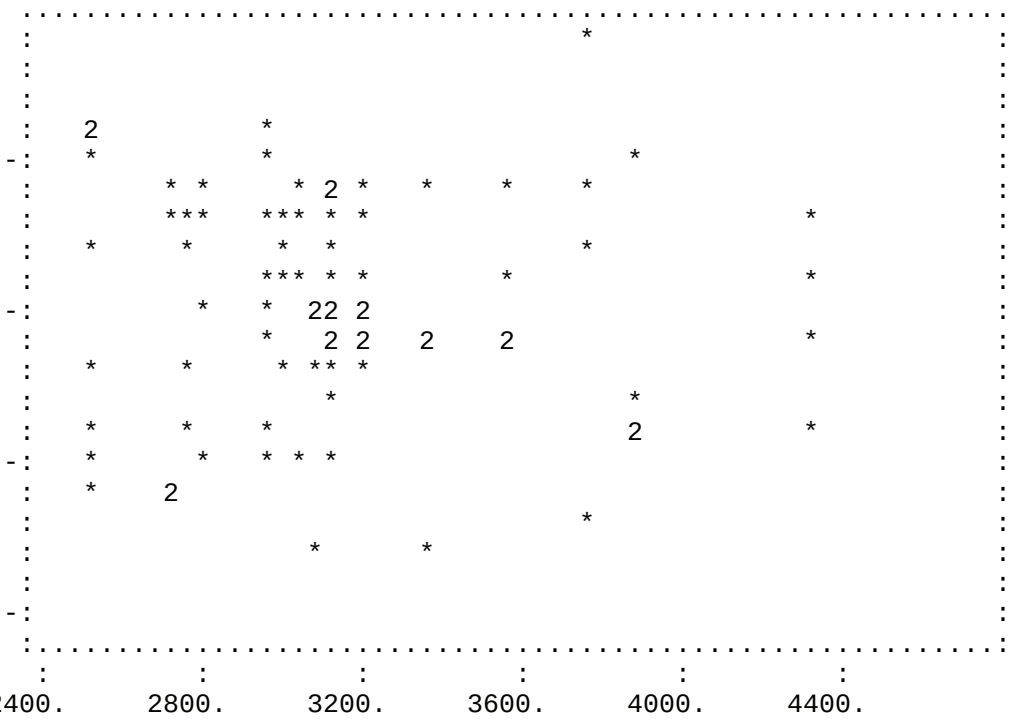
Addendum 3: Spatial analysis of grain yield data, the 2006 and 2007 seasons trials

(A) The 2006 season trials

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/1  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE   1
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                         SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                         DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO VR 2006

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    80

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          80.   1619.       5214.       3174.       731.5

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   20 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)( 13           13)( 14           14)( 15           15)
( 16           16)( 17           17)( 18           18)( 19           19)( 20           20)

    4 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CB          )(  3 CC          )(  4 CD          )(  5 CE          )
(  6 D1          )(  7 DB          )(  8 DC          )(  9 DD          )( 10 DE          )
( 11 DF          )( 12 DG          )( 13 DH          )( 14 Y1          )( 15 Y2          )
( 16 Y3          )( 17 YA          )( 18 YB          )( 19 YC          )( 20 YD          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE   2
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO VR 2006
 Number of non-missing dependent observations:          80
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0

 Message: Relative function convergence

 Final REML criterion:        -424.916207560755026
 Likelihood value -2LogL:      960.105039081510085

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.4372      0.1209       3.616      0.2996E-03
  1 AR1(COLUMN)(2  0.2432      0.1556       1.562      0.1182
```

```
  The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.4078E+06  0.8669E+05   4.704      0.2555E-05
 Asymptotic Covariance Matrix of the Gamma Estimates
                                1           2           3
       1   1 AR1(ROW)(1).   0.146E-01  -0.416E-02   0.462E+04
       2   1 AR1(COLUMN)(  -0.416E-02   0.242E-01   0.197E+04
       3 Dep(1).........   0.462E+04   0.197E+04   0.752E+10
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  60.00        2.868       0.9966E-03

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	2979.	290.1
1	CN$ CB	3116.	297.7
1	CN$ CC	3371.	277.3
1	CN$ CD	3878.	281.0
1	CN$ CE	3013.	281.0
1	CN$ D1	2978.	295.3
1	CN$ DB	3090.	298.6
1	CN$ DC	3129.	285.3
1	CN$ DD	3544.	283.7
1	CN$ DE	3041.	282.8
1	CN$ DF	3207.	286.3
1	CN$ DG	3193.	283.8
1	CN$ DH	2534.	281.8
1	CN$ Y1	2539.	282.9
1	CN$ Y2	2809.	280.3
1	CN$ Y3	2742.	300.5
1	CN$ YA	3138.	286.7
1	CN$ YB	4321.	281.5
1	CN$ YC	3776.	282.5
1	CN$ YD	2731.	286.1

Standard Errors of Differences

Minimum	Mean	Maximum
332.8	369.2	407.4

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

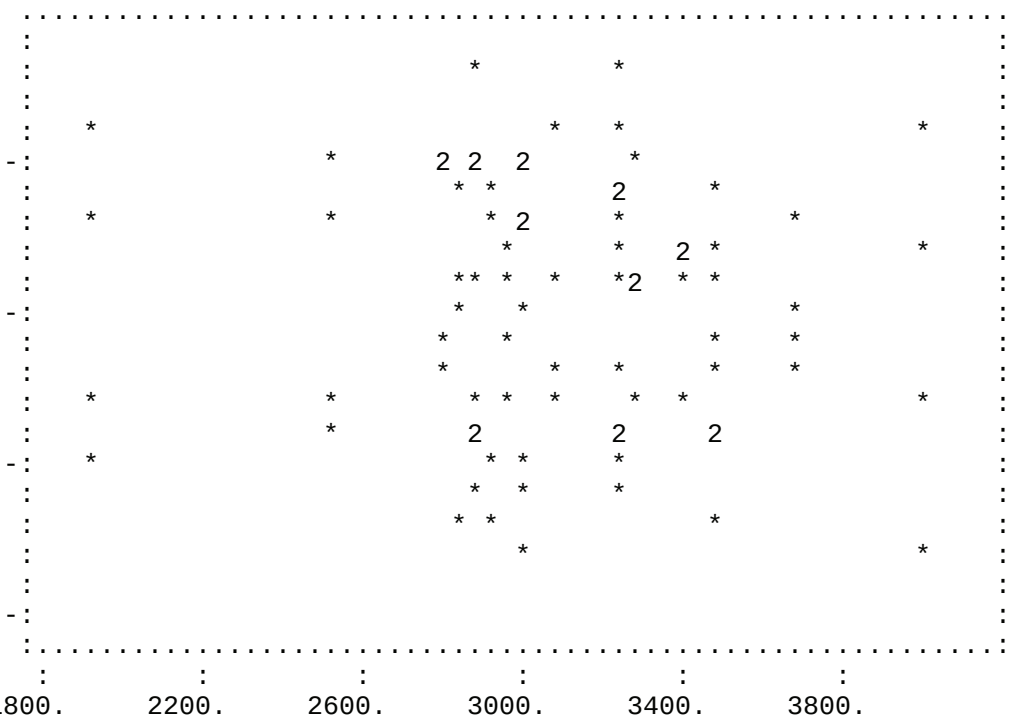
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	2731.	286.1	9.544	0.1371E-20
2	1	CN$	248.5	369.5	0.6727	0.5012
3	1	CN$	385.5	373.7	1.032	0.3023
4	1	CN$	640.0	355.3	1.801	0.7164E-01
5	1	CN$	1148.	379.7	3.023	0.2505E-02
6	1	CN$	281.9	377.1	0.7474	0.4548
7	1	CN$	247.3	367.7	0.6726	0.5012
8	1	CN$	359.2	366.2	0.9810	0.3266
9	1	CN$	398.4	372.0	1.071	0.2841
10	1	CN$	813.4	373.4	2.178	0.2939E-01
11	1	CN$	310.3	360.8	0.8599	0.3899
12	1	CN$	476.6	378.8	1.258	0.2083
13	1	CN$	461.8	381.2	1.211	0.2258
14	1	CN$	-197.0	361.4	-0.5452	0.5856
15	1	CN$	-191.3	381.3	-0.5017	0.6159
16	1	CN$	77.98	338.9	0.2301	0.8180
17	1	CN$	11.31	354.3	0.3194E-01	0.9745
18	1	CN$	407.2	383.9	1.061	0.2887
19	1	CN$	1591.	378.7	4.200	0.2664E-04
20	1	CN$	1045.	379.2	2.757	0.5837E-02

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/2  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE   4
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO LA 2006

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    80

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          80.   680.1       5474.       3089.       1055.

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   20 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)( 13           13)( 14           14)( 15           15)
( 16           16)( 17           17)( 18           18)( 19           19)( 20           20)

    4 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CB          )(  3 CC          )(  4 CD          )(  5 CE          )
(  6 D1          )(  7 DB          )(  8 DC          )(  9 DD          )( 10 DE          )
( 11 DF          )( 12 DG          )( 13 DH          )( 14 Y1          )( 15 Y2          )
( 16 Y3          )( 17 YA          )( 18 YB          )( 19 YC          )( 20 YD          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE   5
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO LA 2006
 Number of non-missing dependent observations:          80
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0

 Message: Relative function convergence

 Final REML criterion:        -435.973380045139095
 Likelihood value -2LogL:      982.219384050278222

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.8018      0.6086E-01   13.17      0.1227E-38
  1 AR1(COLUMN)(2  0.3555      0.1538       2.312      0.2079E-01
```

```
  The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.1225E+07  0.4038E+06   3.033      0.2423E-02
 Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.370E-02   0.195E-02   0.197E+05
       2   1 AR1(COLUMN)(   0.195E-02   0.237E-01   0.246E+05
       3 Dep(1).........   0.197E+05   0.246E+05   0.163E+12
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.
       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  60.00        2.820       0.1185E-02

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	3257.	464.9
1	CN$ CB	1917.	474.0
1	CN$ CC	2849.	473.7
1	CN$ CD	3288.	465.9
1	CN$ CE	2780.	467.5
1	CN$ D1	2539.	477.5
1	CN$ DB	2875.	480.3
1	CN$ DC	3988.	477.6
1	CN$ DD	2903.	474.2
1	CN$ DE	3247.	478.3
1	CN$ DF	2995.	474.4
1	CN$ DG	2862.	471.4
1	CN$ DH	2985.	468.1
1	CN$ Y1	3398.	470.1
1	CN$ Y2	3099.	490.6
1	CN$ Y3	3259.	472.2
1	CN$ YA	3487.	475.5
1	CN$ YB	3482.	470.6
1	CN$ YC	2961.	467.5
1	CN$ YD	3678.	472.0

Standard Errors of Differences

Minimum	Mean	Maximum
284.9	355.7	406.6

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment met
hod

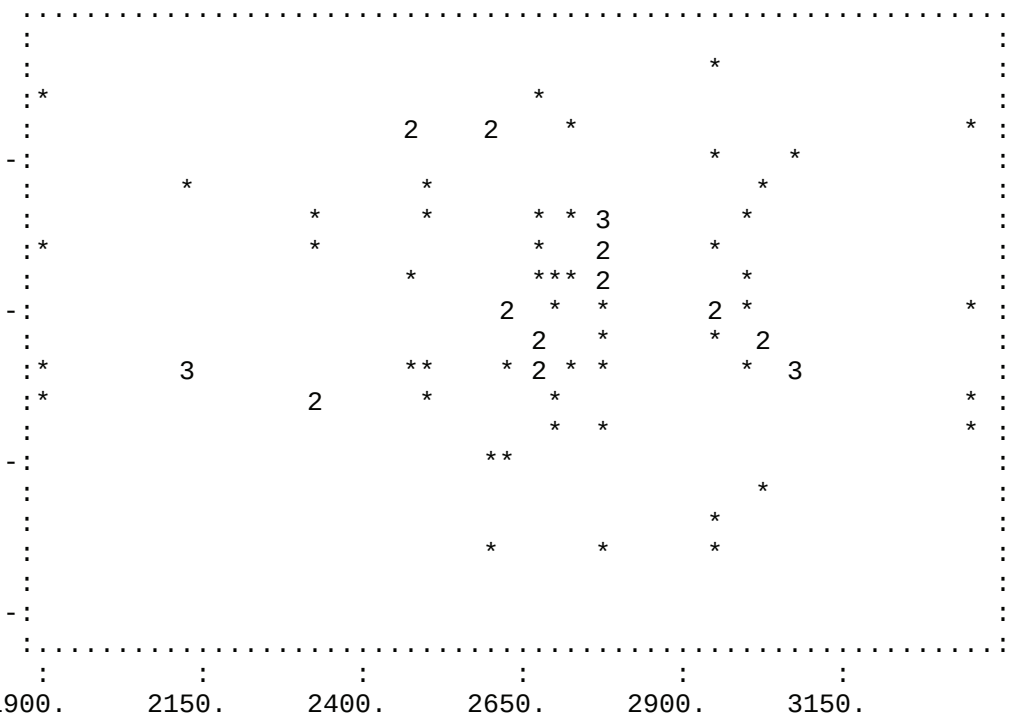
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	3678.	472.0	7.792	0.6593E-14
2	1	CN$	-421.0	362.9	-1.160	0.2460
3	1	CN$	-1760.	361.9	-4.864	0.1152E-05
4	1	CN$	-829.0	375.8	-2.206	0.2737E-01
5	1	CN$	-389.8	349.2	-1.116	0.2642
6	1	CN$	-897.4	348.6	-2.574	0.1004E-01
7	1	CN$	-1139.	356.2	-3.197	0.1387E-02
8	1	CN$	-803.2	394.7	-2.035	0.4183E-01
9	1	CN$	310.6	375.8	0.8266	0.4085
10	1	CN$	-774.5	353.7	-2.190	0.2855E-01
11	1	CN$	-430.7	330.3	-1.304	0.1923
12	1	CN$	-683.2	356.0	-1.919	0.5501E-01
13	1	CN$	-816.1	361.1	-2.260	0.2385E-01
14	1	CN$	-692.8	336.3	-2.060	0.3939E-01
15	1	CN$	-279.9	326.3	-0.8579	0.3909
16	1	CN$	-579.1	364.7	-1.588	0.1123
17	1	CN$	-418.6	368.3	-1.137	0.2557
18	1	CN$	-191.2	366.5	-0.5217	0.6019
19	1	CN$	-195.9	355.2	-0.5517	0.5812
20	1	CN$	-716.5	359.7	-1.992	0.4637E-01

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/3  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE   7
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO ME 2006

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    80

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          80.   1617.       3969.       2710.       480.8

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   20 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)( 13           13)( 14           14)( 15           15)
( 16           16)( 17           17)( 18           18)( 19           19)( 20           20)

    4 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CB          )(  3 CC          )(  4 CD          )(  5 CE          )
(  6 D1          )(  7 DB          )(  8 DC          )(  9 DD          )( 10 DE          )
( 11 DF          )( 12 DG          )( 13 DH          )( 14 Y1          )( 15 Y2          )
( 16 Y3          )( 17 YA          )( 18 YB          )( 19 YC          )( 20 YD          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE   8
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO ME 2006
 Number of non-missing dependent observations:          80
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -387.203885355540308
 Likelihood value -2LogL:      884.680394671080649

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.3696      0.1082       3.416      0.6347E-03
  1 AR1(COLUMN)(2  0.6348      0.1096       5.792      0.6967E-08
```

```
 The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.1450E+06  0.3521E+05   4.120      0.3792E-04
Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.117E-01  -0.117E-02    933.
       2   1 AR1(COLUMN)(  -0.117E-02   0.120E-01   0.226E+04
       3 Dep(1).........    933.       0.226E+04   0.124E+10
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  60.00        6.906       0.3785E-08

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	3065.	152.1
1	CN$ CB	1904.	154.5
1	CN$ CC	2512.	157.5
1	CN$ CD	2116.	173.4
1	CN$ CE	3019.	156.3
1	CN$ D1	2958.	152.4
1	CN$ DB	2775.	151.0
1	CN$ DC	2991.	154.7
1	CN$ DD	2703.	159.9
1	CN$ DE	2951.	155.4
1	CN$ DF	2764.	156.1
1	CN$ DG	2318.	156.1
1	CN$ DH	2675.	155.1
1	CN$ Y1	2784.	150.2
1	CN$ Y2	2717.	156.2
1	CN$ Y3	3345.	157.0
1	CN$ YA	2666.	150.9
1	CN$ YB	2636.	156.0
1	CN$ YC	2590.	152.4
1	CN$ YD	2480.	169.9

Standard Errors of Differences

Minimum	Mean	Maximum
147.6	180.6	213.2

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment met
hod

The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	2480.	169.9	14.60	0.2923E-47
2	1	CN$	584.6	198.7	2.942	0.3265E-02
3	1	CN$	-576.4	198.0	-2.911	0.3604E-02
4	1	CN$	32.19	179.6	0.1793	0.8577
5	1	CN$	-364.4	185.2	-1.967	0.4917E-01
6	1	CN$	538.7	202.9	2.655	0.7921E-02
7	1	CN$	477.9	185.2	2.581	0.9857E-02
8	1	CN$	295.3	192.1	1.537	0.1243
9	1	CN$	511.0	190.8	2.679	0.7393E-02
10	1	CN$	223.2	209.6	1.065	0.2868
11	1	CN$	471.0	199.7	2.358	0.1838E-01
12	1	CN$	283.7	197.1	1.440	0.1500
13	1	CN$	-161.8	176.2	-0.9180	0.3586
14	1	CN$	195.4	200.1	0.9768	0.3287
15	1	CN$	304.4	180.7	1.685	0.9205E-01
16	1	CN$	236.5	187.4	1.262	0.2069
17	1	CN$	865.5	183.4	4.719	0.2374E-05
18	1	CN$	186.1	195.6	0.9515	0.3414
19	1	CN$	156.0	201.6	0.7739	0.4390
20	1	CN$	110.3	196.9	0.5602	0.5753

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/4  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  10
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO RO 2006

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    80

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          80.   3945.       7144.       5564.       686.1

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   20 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)( 13           13)( 14           14)( 15           15)
( 16           16)( 17           17)( 18           18)( 19           19)( 20           20)

    4 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CB          )(  3 CC          )(  4 CD          )(  5 CE          )
(  6 D1          )(  7 DB          )(  8 DC          )(  9 DD          )( 10 DE          )
( 11 DF          )( 12 DG          )( 13 DH          )( 14 Y1          )( 15 Y2          )
( 16 Y3          )( 17 YA          )( 18 YB          )( 19 YC          )( 20 YD          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  11
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO RO 2006
 Number of non-missing dependent observations:          80
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -421.343823343138070
 Likelihood value -2LogL:      952.960270646276172

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.4777      0.1058       4.517      0.6280E-05
  1 AR1(COLUMN)(2  0.3064      0.1618       1.894      0.5824E-01
```

The scale parameters

Dep.	Sigma_Squared	Std. Error	Z	Pr > |Z|
Dep(1)	0.3865E+06	0.8614E+05	4.487	0.7240E-05

Asymptotic Covariance Matrix of the Gamma Estimates

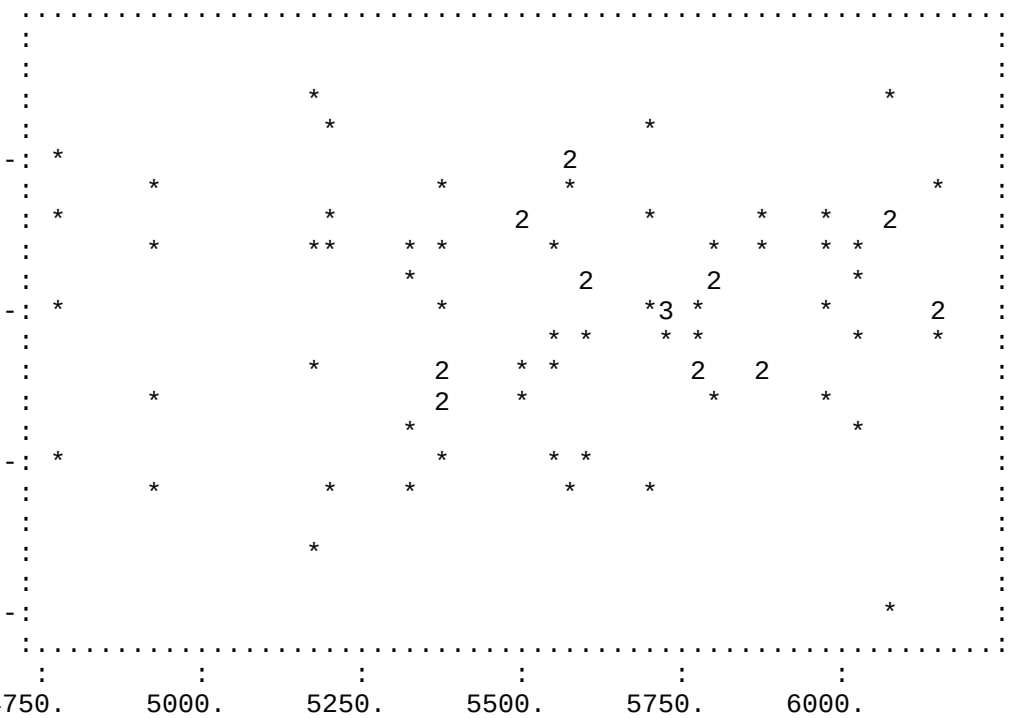
		1	2	3
1	1 AR1(ROW)(1).	0.112E-01	-0.885E-03	0.429E+04
2	1 AR1(COLUMN)(	-0.885E-03	0.262E-01	0.398E+04
3	Dep(1).....	0.429E+04	0.398E+04	0.742E+10

Warning: Denominator degrees of freedom estimates do not account for measurement error parameters.

ANOVA Table for Sequentially Deleted Fixed Effects
Denominator Degrees of Freedom: Containment method

Dep	Effect	DFNum	DFDen	F - Statistic	P > |F|
1	CN$	19	60.00	2.627	0.2359E-02

Plot of residuals against predicted values



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	6138.	265.1
1	CN$ CB	4780.	267.3
1	CN$ CC	5320.	275.8
1	CN$ CD	5562.	279.7
1	CN$ CE	5592.	272.2
1	CN$ D1	5370.	276.6
1	CN$ DB	5794.	271.9
1	CN$ DC	4914.	274.1
1	CN$ DD	5174.	269.4
1	CN$ DE	5506.	264.6
1	CN$ DF	5376.	273.3
1	CN$ DG	5202.	269.3
1	CN$ DH	5886.	267.1
1	CN$ Y1	5965.	272.5
1	CN$ Y2	5736.	269.3
1	CN$ Y3	5699.	273.9
1	CN$ YA	5568.	274.8
1	CN$ YB	6076.	270.1
1	CN$ YC	6037.	285.2
1	CN$ YD	5775.	270.2

Standard Errors of Differences

Minimum	Mean	Maximum
294.0	339.1	376.1

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	5775.	270.2	21.37	0.2448-100
2	1	CN$	362.9	340.0	1.067	0.2858
3	1	CN$	-995.6	310.6	-3.205	0.1349E-02
4	1	CN$	-455.4	334.7	-1.361	0.1736
5	1	CN$	-213.0	362.1	-0.5884	0.5562
6	1	CN$	-183.6	345.4	-0.5314	0.5952
7	1	CN$	-405.6	334.4	-1.213	0.2252
8	1	CN$	18.83	326.9	0.5761E-01	0.9541
9	1	CN$	-861.2	350.5	-2.457	0.1401E-01
10	1	CN$	-601.1	341.8	-1.759	0.7865E-01
11	1	CN$	-268.9	312.2	-0.8611	0.3892
12	1	CN$	-398.9	346.6	-1.151	0.2498
13	1	CN$	-573.6	322.5	-1.779	0.7529E-01
14	1	CN$	110.7	325.3	0.3403	0.7336
15	1	CN$	189.5	351.0	0.5400	0.5892
16	1	CN$	-38.69	324.4	-0.1193	0.9051
17	1	CN$	-75.95	322.1	-0.2358	0.8136
18	1	CN$	-206.8	347.9	-0.5944	0.5522
19	1	CN$	300.8	353.8	0.8503	0.3952
20	1	CN$	262.1	340.3	0.7702	0.4412

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/5  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  13
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO TY 2006

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    80

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          80.   2491.       5659.       4091.       567.2

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   20 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)( 13           13)( 14           14)( 15           15)
( 16           16)( 17           17)( 18           18)( 19           19)( 20           20)

    4 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CB          )(  3 CC          )(  4 CD          )(  5 CE          )
(  6 D1          )(  7 DB          )(  8 DC          )(  9 DD          )( 10 DE          )
( 11 DF          )( 12 DG          )( 13 DH          )( 14 Y1          )( 15 Y2          )
( 16 Y3          )( 17 YA          )( 18 YB          )( 19 YC          )( 20 YD          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  14
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO TY 2006
 Number of non-missing dependent observations:          80
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                       1-  2   product   0     1      1
   AR1(ROW)                     1-  1       AR1   0     1      1
   AR1(COLUMN)                  2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -400.555301643308496
 Likelihood value -2LogL:      911.383227246617025

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.4753      0.1129       4.210      0.2555E-04
  1 AR1(COLUMN)(2 -0.1101E-01  0.2024     -0.5441E-01  0.9566
```

The scale parameters

Dep.	Sigma_Squared	Std. Error	Z	Pr > |Z|
Dep(1)	0.1820E+06	0.3959E+05	4.598	0.4267E-05

Asymptotic Covariance Matrix of the Gamma Estimates

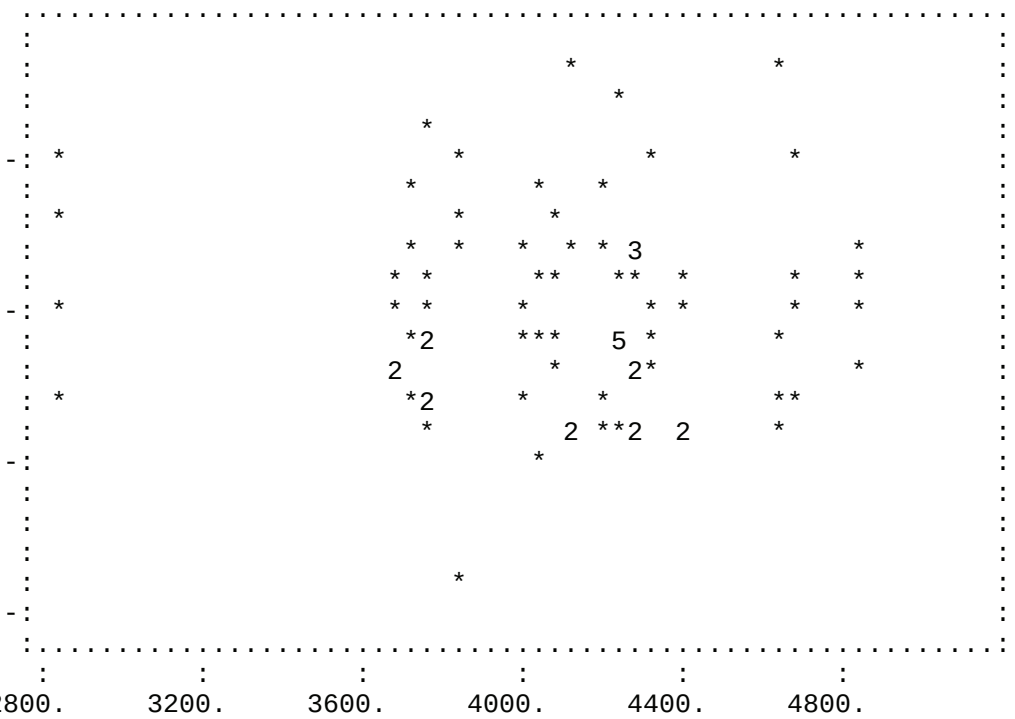
		1	2	3
1	1 AR1(ROW)(1).	0.127E-01	-0.166E-02	0.236E+04
2	1 AR1(COLUMN)(	-0.166E-02	0.410E-01	-267.
3	Dep(1).....	0.236E+04	-267.	0.157E+10

Warning: Denominator degrees of freedom estimates do not account for measurement error parameters.

ANOVA Table for Sequentially Deleted Fixed Effects
Denominator Degrees of Freedom: Containment method

Dep	Effect	DFNum	DFDen	F - Statistic	P > |F|
1	CN$	19	60.00	6.386	0.1516E-07

Plot of residuals against predicted values



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	4190.	188.2
1	CN$ CB	4104.	184.1
1	CN$ CC	4310.	183.0
1	CN$ CD	2828.	184.4
1	CN$ CE	3984.	183.0
1	CN$ D1	3670.	184.6
1	CN$ DB	3831.	186.0
1	CN$ DC	4082.	186.2
1	CN$ DD	3766.	184.5
1	CN$ DE	4241.	188.6
1	CN$ DF	4024.	187.5
1	CN$ DG	3773.	184.2
1	CN$ DH	3709.	184.2
1	CN$ Y1	4282.	185.7
1	CN$ Y2	4648.	182.8
1	CN$ Y3	4411.	186.9
1	CN$ YA	4259.	187.5
1	CN$ YB	4668.	183.1
1	CN$ YC	4858.	183.0
1	CN$ YD	4295.	186.5

Standard Errors of Differences

Minimum	Mean	Maximum
223.8	244.8	257.9

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

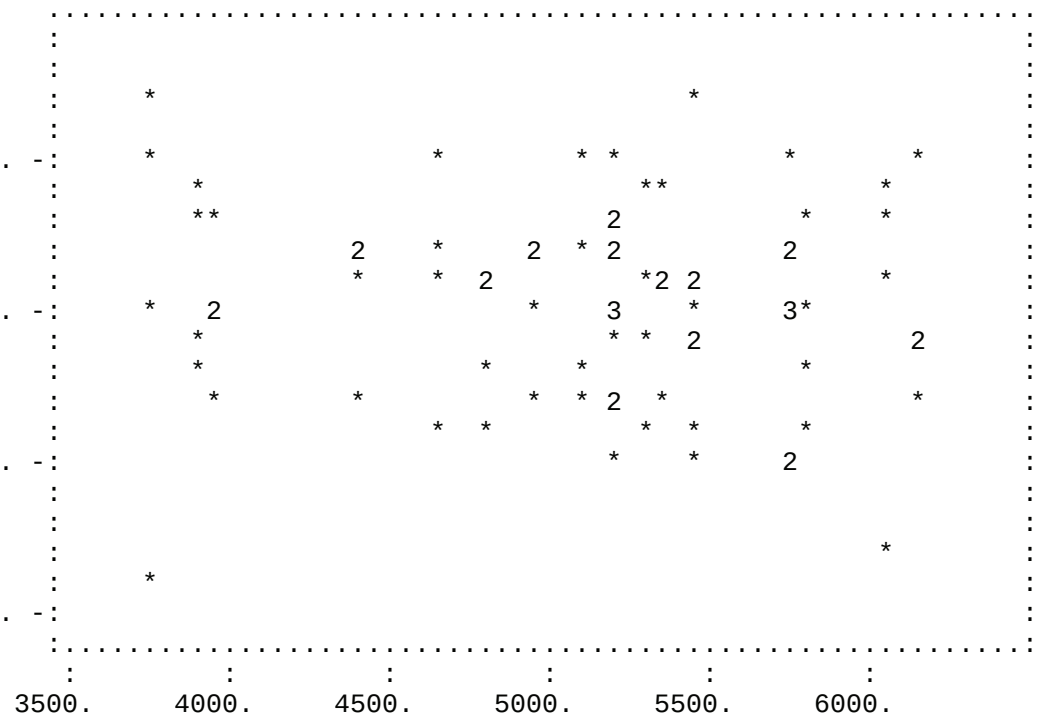
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	4295.	186.5	23.03	0.2161-116
2	1	CN$	-104.3	256.2	-0.4071	0.6839
3	1	CN$	-190.4	250.1	-0.7615	0.4464
4	1	CN$	14.99	236.8	0.6331E-01	0.9495
5	1	CN$	-1466.	250.7	-5.850	0.4924E-08
6	1	CN$	-310.4	236.2	-1.314	0.1889
7	1	CN$	-624.7	223.8	-2.791	0.5257E-02
8	1	CN$	-464.1	248.9	-1.865	0.6222E-01
9	1	CN$	-212.9	252.5	-0.8433	0.3990
10	1	CN$	-528.3	251.3	-2.102	0.3554E-01
11	1	CN$	-53.37	256.9	-0.2077	0.8355
12	1	CN$	-271.0	252.1	-1.075	0.2824
13	1	CN$	-521.7	250.8	-2.080	0.3751E-01
14	1	CN$	-585.3	248.5	-2.355	0.1851E-01
15	1	CN$	-12.87	250.4	-0.5137E-01	0.9590
16	1	CN$	353.1	235.6	1.499	0.1340
17	1	CN$	116.3	227.9	0.5105	0.6097
18	1	CN$	-35.70	249.7	-0.1430	0.8863
19	1	CN$	373.7	234.3	1.595	0.1108
20	1	CN$	563.5	243.7	2.313	0.2075E-01

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/6  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  16
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO NA 2006

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    80

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          80.   2346.       6934.       5141.       829.8

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   20 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)( 13           13)( 14           14)( 15           15)
( 16           16)( 17           17)( 18           18)( 19           19)( 20           20)

    4 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(

   20 CODES:(Number  Label) for Variable: CN$

(  1 CA          )(  2 CB          )(  3 CC          )(  4 CD          )(  5 CE          )
(  6 D1          )(  7 DB          )(  8 DC          )(  9 DD          )( 10 DE          )
( 11 DF          )( 12 DG          )( 13 DH          )( 14 Y1          )( 15 Y2          )
( 16 Y3          )( 17 YA          )( 18 YB          )( 19 YC          )( 20 YD          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  17
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO NA 2006
 Number of non-missing dependent observations:          80
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -421.818268383729333
 Likelihood value -2LogL:      953.909160727458698

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.1735      0.1344       1.290      0.1969
  1 AR1(COLUMN)(2  0.3863      0.1408       2.743      0.6085E-02
```

```
The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.3313E+06  0.6680E+05   4.961      0.7030E-06
Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.181E-01   0.146E-02   0.180E+04
       2   1 AR1(COLUMN)(   0.146E-02   0.198E-01   0.382E+04
       3 Dep(1).........   0.180E+04   0.382E+04   0.446E+10
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  60.00        8.117       0.1845E-09

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	5201.	261.9
1	CN$ CB	4379.	262.3
1	CN$ CC	5798.	263.7
1	CN$ CD	3939.	262.4
1	CN$ CE	4645.	267.8
1	CN$ D1	5446.	266.3
1	CN$ DB	5474.	289.9
1	CN$ DC	5280.	287.8
1	CN$ DD	4954.	260.6
1	CN$ DE	5121.	262.7
1	CN$ DF	4806.	264.3
1	CN$ DG	3914.	265.0
1	CN$ DH	5218.	287.4
1	CN$ Y1	5762.	266.4
1	CN$ Y2	6139.	258.6
1	CN$ Y3	6052.	291.2
1	CN$ YA	5325.	262.5
1	CN$ YB	3729.	264.2
1	CN$ YC	5768.	262.8
1	CN$ YD	5181.	264.2

Standard Errors of Differences

Minimum	Mean	Maximum
318.0	362.0	406.5

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

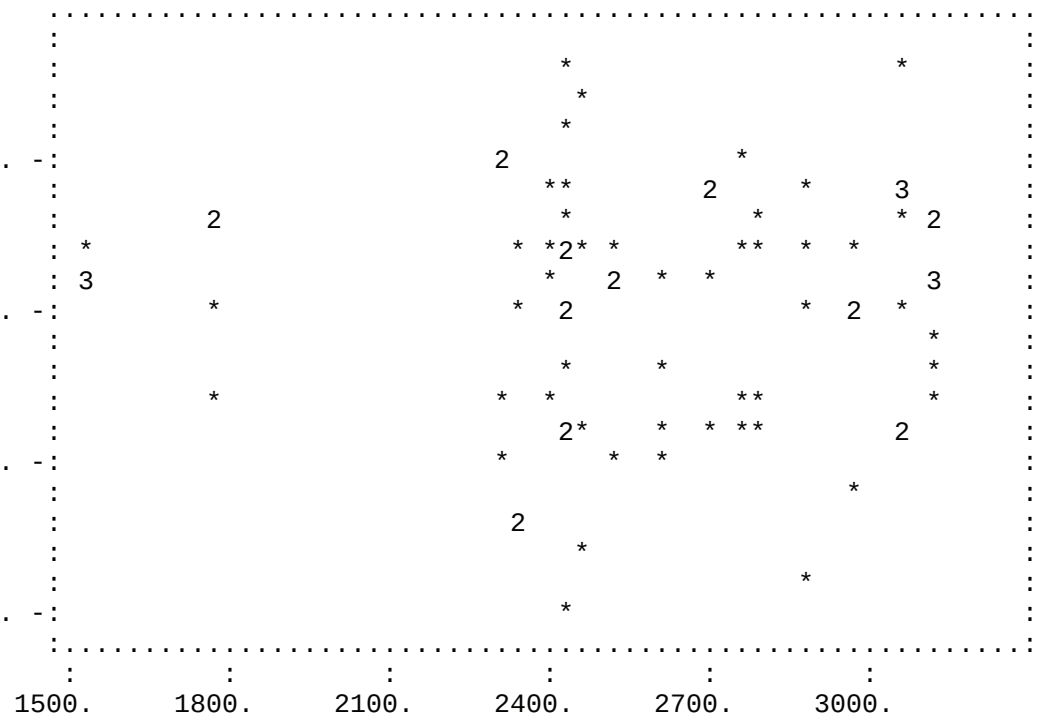
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	5181.	264.2	19.61	0.1371E-84
2	1	CN$	20.42	352.6	0.5790E-01	0.9538
3	1	CN$	-802.2	360.7	-2.224	0.2614E-01
4	1	CN$	617.5	350.7	1.761	0.7825E-01
5	1	CN$	-1242.	354.4	-3.505	0.4559E-03
6	1	CN$	-535.9	340.6	-1.574	0.1156
7	1	CN$	265.4	366.4	0.7242	0.4690
8	1	CN$	292.8	387.8	0.7550	0.4503
9	1	CN$	99.42	383.9	0.2590	0.7957
10	1	CN$	-226.3	353.8	-0.6395	0.5225
11	1	CN$	-59.39	360.3	-0.1648	0.8691
12	1	CN$	-374.7	352.0	-1.065	0.2871
13	1	CN$	-1267.	366.4	-3.457	0.5461E-03
14	1	CN$	37.04	362.3	0.1022	0.9186
15	1	CN$	581.4	347.4	1.674	0.9418E-01
16	1	CN$	958.1	349.6	2.741	0.6128E-02
17	1	CN$	871.6	338.9	2.572	0.1011E-01
18	1	CN$	144.6	362.8	0.3986	0.6902
19	1	CN$	-1451.	364.9	-3.977	0.6969E-04
20	1	CN$	587.1	363.2	1.616	0.1060

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/7  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  19
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO MA 2006

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    80

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          80.   1501.       3671.       2596.       518.3

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   20 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)( 13           13)( 14           14)( 15           15)
( 16           16)( 17           17)( 18           18)( 19           19)( 20           20)

    4 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(

   20 CODES:(Number  Label) for Variable: CN$
(  1 BA          )(  2 BB          )(  3 BC          )(  4 BD          )(  5 BE          )
(  6 BF          )(  7 BG          )(  8 BH          )(  9 BI          )( 10 BJ          )
( 11 BK          )( 12 BL          )( 13 BM          )( 14 BN          )( 15 BO          )
( 16 CA          )( 17 CB          )( 18 CC          )( 19 CD          )( 20 CE          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  20
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO MA 2006
 Number of non-missing dependent observations:          80
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -384.584924195157271
 Likelihood value -2LogL:      879.442472350314574

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.3076      0.1280       2.403      0.1628E-01
  1 AR1(COLUMN)(2  0.4711      0.1315       3.581      0.3423E-03
```

```
The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.1096E+06  0.2375E+05   4.614      0.3957E-05
Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.164E-01  -0.192E-02    795.
       2   1 AR1(COLUMN)(  -0.192E-02   0.173E-01   0.136E+04
       3 Dep(1).........    795.       0.136E+04   0.564E+09
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  60.00        11.72       0.9350E-13

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ BA	2454.	146.0
1	CN$ BB	2302.	142.1
1	CN$ BC	2770.	143.2
1	CN$ BD	2341.	141.5
1	CN$ BE	2781.	155.9
1	CN$ BF	2528.	147.5
1	CN$ BG	2444.	146.2
1	CN$ BH	2692.	146.2
1	CN$ BI	3046.	144.5
1	CN$ BJ	3061.	142.0
1	CN$ BK	2429.	157.1
1	CN$ BL	2441.	158.2
1	CN$ BM	2413.	145.0
1	CN$ BN	3125.	145.1
1	CN$ BO	2976.	144.8
1	CN$ CA	3133.	147.6
1	CN$ CB	1766.	145.4
1	CN$ CC	2602.	145.6
1	CN$ CD	1520.	144.3
1	CN$ CE	2872.	153.9

Standard Errors of Differences

Minimum	Mean	Maximum
161.6	188.3	209.1

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

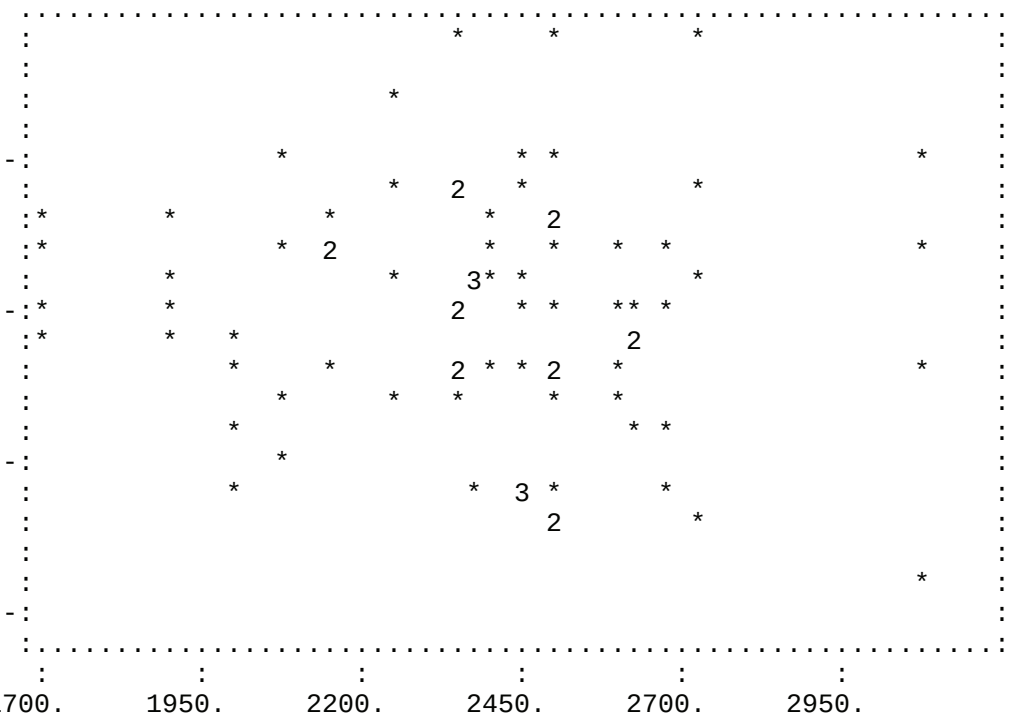
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	2872.	153.9	18.66	0.9599E-77
2	1	CN$	-418.7	184.1	-2.274	0.2297E-01
3	1	CN$	-570.5	181.0	-3.153	0.1617E-02
4	1	CN$	-101.9	193.8	-0.5258	0.5990
5	1	CN$	-531.0	192.8	-2.755	0.5876E-02
6	1	CN$	-91.22	190.5	-0.4790	0.6320
7	1	CN$	-344.6	202.1	-1.705	0.8824E-01
8	1	CN$	-428.4	199.9	-2.143	0.3208E-01
9	1	CN$	-179.8	201.1	-0.8942	0.3712
10	1	CN$	173.4	198.5	0.8733	0.3825
11	1	CN$	189.1	198.7	0.9518	0.3412
12	1	CN$	-442.9	199.8	-2.217	0.2662E-01
13	1	CN$	-431.5	209.1	-2.063	0.3908E-01
14	1	CN$	-458.8	193.5	-2.372	0.1770E-01
15	1	CN$	252.5	189.3	1.334	0.1822
16	1	CN$	103.5	189.9	0.5452	0.5856
17	1	CN$	260.6	199.6	1.305	0.1917
18	1	CN$	-1106.	196.3	-5.635	0.1749E-07
19	1	CN$	-270.3	179.6	-1.505	0.1324
20	1	CN$	-1352.	176.7	-7.651	0.1998E-13

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/8  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  22
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO MB 2006

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    80

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          80.   1290.       3849.       2366.       539.1

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   20 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)( 13           13)( 14           14)( 15           15)
( 16           16)( 17           17)( 18           18)( 19           19)( 20           20)

    4 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(

   20 CODES:(Number  Label) for Variable: CN$
(  1 BP          )(  2 BQ          )(  3 BR          )(  4 BS          )(  5 BT          )
(  6 BU          )(  7 BV          )(  8 BW          )(  9 BX          )( 10 BY          )
( 11 BZ          )( 12 CA          )( 13 CB          )( 14 CC          )( 15 CD          )
( 16 CE          )( 17 EA          )( 18 UA          )( 19 UB          )( 20 UC          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  23
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO MB 2006
 Number of non-missing dependent observations:          80
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -394.443328943878555
 Likelihood value -2LogL:      899.159281847757143

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.3296      0.1241       2.657      0.7892E-02
  1 AR1(COLUMN)(2  0.7517      0.7856E-01   9.569      0.1084E-20
```

```
The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.2230E+06  0.5589E+05   3.990      0.6607E-04
Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.154E-01  -0.274E-02    652.
       2   1 AR1(COLUMN)(  -0.274E-02   0.617E-02   0.274E+04
       3 Dep(1).........    652.       0.274E+04   0.312E+10
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  60.00        8.282       0.1247E-09

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ BP	2491.	180.6
1	CN$ BQ	2345.	176.3
1	CN$ BR	2361.	176.9
1	CN$ BS	2010.	174.3
1	CN$ BT	2629.	192.2
1	CN$ BU	2157.	181.6
1	CN$ BV	2507.	181.0
1	CN$ BW	2510.	180.8
1	CN$ BX	2076.	176.6
1	CN$ BY	2257.	174.2
1	CN$ BZ	2410.	195.4
1	CN$ CA	3075.	180.6
1	CN$ CB	1903.	177.6
1	CN$ CC	2673.	179.3
1	CN$ CD	1711.	177.2
1	CN$ CE	2451.	189.1
1	CN$ EA	2718.	177.8
1	CN$ UA	2451.	195.3
1	CN$ UB	2590.	179.0
1	CN$ UC	2377.	178.5

Standard Errors of Differences

Minimum	Mean	Maximum
156.7	194.6	226.3

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

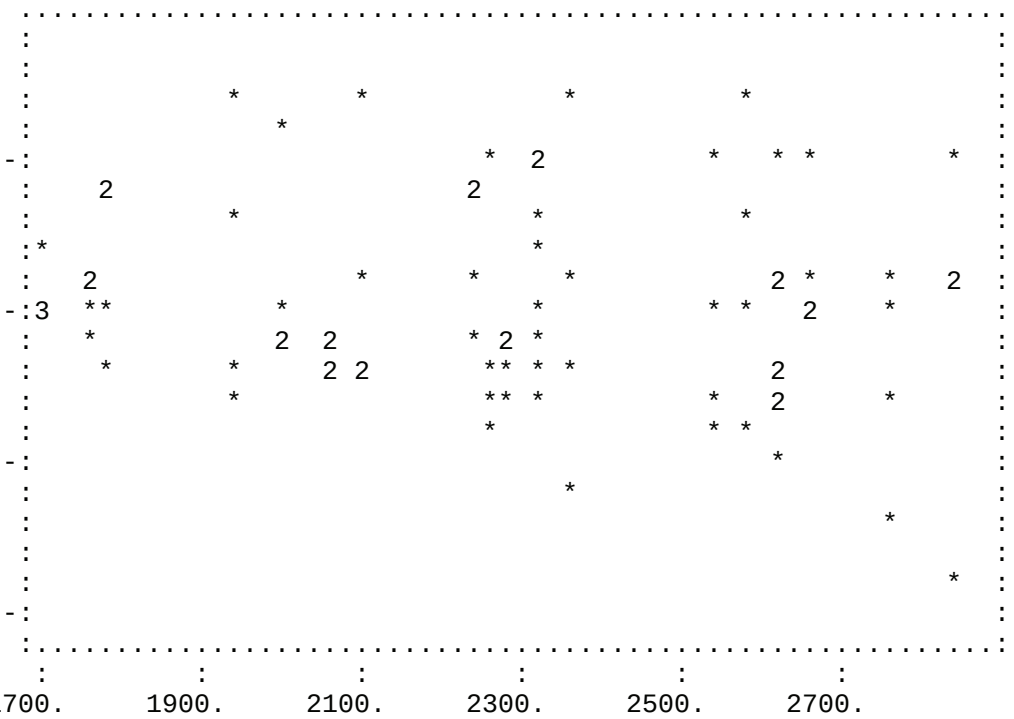
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	2377.	178.5	13.32	0.1827E-39
2	1	CN$	114.1	187.0	0.6101	0.5418
3	1	CN$	-31.70	194.8	-0.1628	0.8707
4	1	CN$	-15.64	167.8	-0.9320E-01	0.9257
5	1	CN$	-367.5	176.9	-2.077	0.3776E-01
6	1	CN$	252.0	202.3	1.245	0.2129
7	1	CN$	-220.4	194.1	-1.136	0.2560
8	1	CN$	130.4	188.9	0.6904	0.4900
9	1	CN$	132.9	204.0	0.6513	0.5149
10	1	CN$	-301.4	197.2	-1.529	0.1264
11	1	CN$	-119.6	192.4	-0.6216	0.5342
12	1	CN$	32.81	216.5	0.1516	0.8795
13	1	CN$	698.2	206.0	3.389	0.7023E-03
14	1	CN$	-474.1	197.0	-2.407	0.1609E-01
15	1	CN$	296.2	161.7	1.832	0.6698E-01
16	1	CN$	-665.6	199.5	-3.337	0.8472E-03
17	1	CN$	73.88	196.3	0.3764	0.7067
18	1	CN$	340.9	176.9	1.927	0.5399E-01
19	1	CN$	73.59	203.8	0.3611	0.7181
20	1	CN$	212.5	187.9	1.131	0.2580

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/9  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  25
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO MC 2006

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    80

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          80.   1486.       3426.       2302.       483.0

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   20 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)( 13           13)( 14           14)( 15           15)
( 16           16)( 17           17)( 18           18)( 19           19)( 20           20)

    4 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CB          )(  3 CC          )(  4 CD          )(  5 CE          )
(  6 UD          )(  7 UE          )(  8 UF          )(  9 UG          )( 10 UH          )
( 11 UI          )( 12 UJ          )( 13 UK          )( 14 UM          )( 15 UN          )
( 16 UO          )( 17 UP          )( 18 UQ          )( 19 UR          )( 20 US          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  26
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO MC 2006
 Number of non-missing dependent observations:          80
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -390.533815568266391
 Likelihood value -2LogL:      891.340255096532815

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.4699      0.1135       4.139      0.3488E-04
  1 AR1(COLUMN)(2  0.6113      0.1126       5.431      0.5598E-07
```

```
The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.1754E+06  0.4334E+05   4.048      0.5175E-04
Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.129E-01  -0.265E-02   0.168E+04
       2   1 AR1(COLUMN)(  -0.265E-02   0.127E-01   0.239E+04
       3 Dep(1).........   0.168E+04   0.239E+04   0.188E+10
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  60.00        9.311       0.1211E-10

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	2627.	175.7
1	CN$ CB	1760.	172.0
1	CN$ CC	2250.	172.8
1	CN$ CD	1709.	171.4
1	CN$ CE	2330.	179.6
1	CN$ UD	2534.	174.5
1	CN$ UE	2358.	168.1
1	CN$ UF	2060.	169.2
1	CN$ UG	2767.	167.7
1	CN$ UH	2285.	181.5
1	CN$ UI	1771.	175.7
1	CN$ UJ	2841.	174.1
1	CN$ UK	2571.	173.8
1	CN$ UM	1936.	171.2
1	CN$ UN	2323.	167.4
1	CN$ UO	2670.	184.2
1	CN$ UP	2618.	186.1
1	CN$ UQ	2230.	171.9
1	CN$ UR	1995.	171.0
1	CN$ US	2102.	171.4

Standard Errors of Differences

Minimum	Mean	Maximum
154.9	190.4	215.3

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

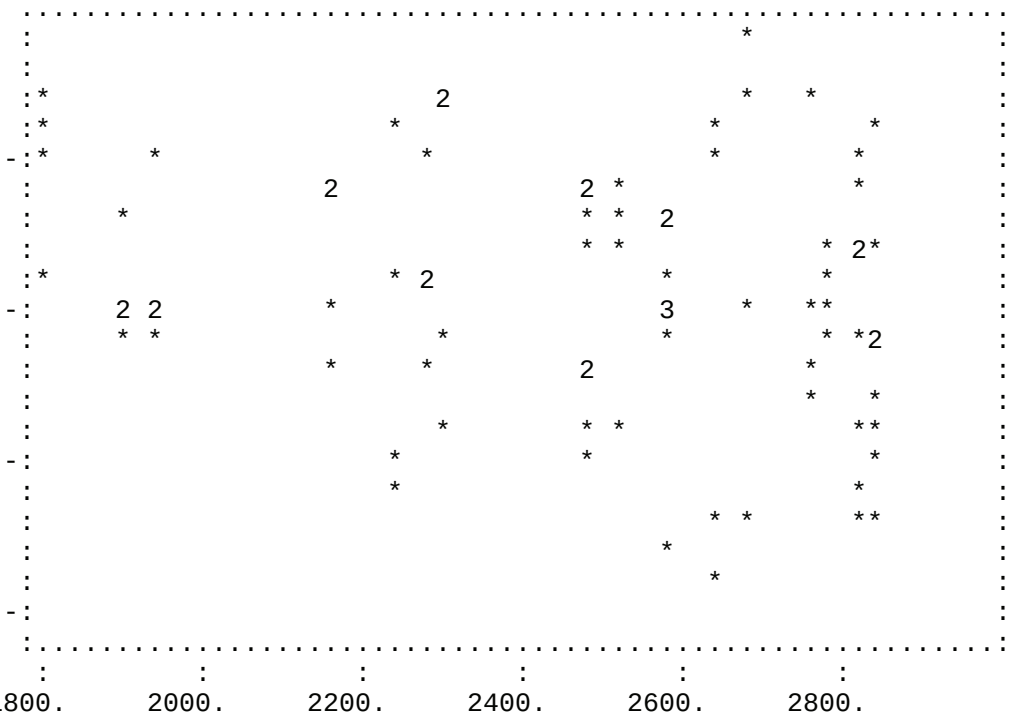
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	2102.	171.4	12.26	0.1484E-33
2	1	CN$	525.0	204.7	2.565	0.1031E-01
3	1	CN$	-341.2	195.6	-1.744	0.8111E-01
4	1	CN$	148.9	163.4	0.9110	0.3623
5	1	CN$	-392.5	195.7	-2.006	0.4487E-01
6	1	CN$	228.1	188.6	1.210	0.2264
7	1	CN$	432.5	185.3	2.335	0.1956E-01
8	1	CN$	256.8	187.3	1.371	0.1703
9	1	CN$	-41.38	164.7	-0.2512	0.8017
10	1	CN$	665.5	176.2	3.776	0.1593E-03
11	1	CN$	183.3	194.4	0.9429	0.3457
12	1	CN$	-330.2	193.8	-1.704	0.8835E-01
13	1	CN$	739.8	189.1	3.913	0.9107E-04
14	1	CN$	469.2	198.3	2.367	0.1796E-01
15	1	CN$	-165.3	194.7	-0.8491	0.3958
16	1	CN$	222.0	189.1	1.173	0.2406
17	1	CN$	568.4	206.7	2.750	0.5964E-02
18	1	CN$	516.5	196.7	2.626	0.8629E-02
19	1	CN$	128.6	174.7	0.7361	0.4617
20	1	CN$	-106.5	181.2	-0.5877	0.5567

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/10  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  28
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO MD 2006

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    80

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          80.   1556.       3804.       2517.       528.5

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   20 CODES:(Number  Label) for Variable: ROW
(  1           1)(  2           2)(  3           3)(  4           4)(  5           5)
(  6           6)(  7           7)(  8           8)(  9           9)( 10          10)
( 11          11)( 12          12)( 13          13)( 14          14)( 15          15)
( 16          16)( 17          17)( 18          18)( 19          19)( 20          20)

    4 CODES:(Number  Label) for Variable: COLUMN
(  1           1)(  2           2)(  3           3)(  4           4)(

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA         )(  2 CB         )(  3 CC         )(  4 CD         )(  5 CE         )
(  6 UT         )(  7 UU         )(  8 UV         )(  9 UW         )( 10 UX         )
( 11 UY         )( 12 UZ         )( 13 ZA         )( 14 ZB         )( 15 ZC         )
( 16 ZD         )( 17 ZE         )( 18 ZF         )( 19 ZG         )( 20 ZH         )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  29
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO MD 2006
 Number of non-missing dependent observations:          80
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -392.519561856171094
 Likelihood value -2LogL:      895.311747672342221

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.5969      0.9258E-01   6.447      0.1139E-09
  1 AR1(COLUMN)(2  0.8263      0.5886E-01   14.04      0.9128E-44
```

```
The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.3357E+06  0.1081E+06   3.105      0.1906E-02
Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.857E-02   0.767E-03   0.540E+04
       2   1 AR1(COLUMN)(   0.767E-03   0.346E-02   0.441E+04
       3 Dep(1).........   0.540E+04   0.441E+04   0.117E+11
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  60.00        8.467       0.8106E-10

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	2635.	241.0
1	CN$ CB	2236.	238.1
1	CN$ CC	2282.	239.9
1	CN$ CD	1934.	238.6
1	CN$ CE	2481.	243.2
1	CN$ UT	2520.	242.2
1	CN$ UU	2586.	237.1
1	CN$ UV	2588.	236.9
1	CN$ UW	2846.	236.6
1	CN$ UX	2479.	244.1
1	CN$ UY	2683.	241.9
1	CN$ UZ	2769.	241.4
1	CN$ ZA	2777.	241.0
1	CN$ ZB	2811.	238.2
1	CN$ ZC	2823.	235.7
1	CN$ ZD	2831.	247.5
1	CN$ ZE	1898.	248.4
1	CN$ ZF	1798.	238.9
1	CN$ ZG	2153.	237.9
1	CN$ ZH	2310.	238.6

Standard Errors of Differences

Minimum	Mean	Maximum
124.0	162.6	190.5

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

The Regression Coefficient Estimates

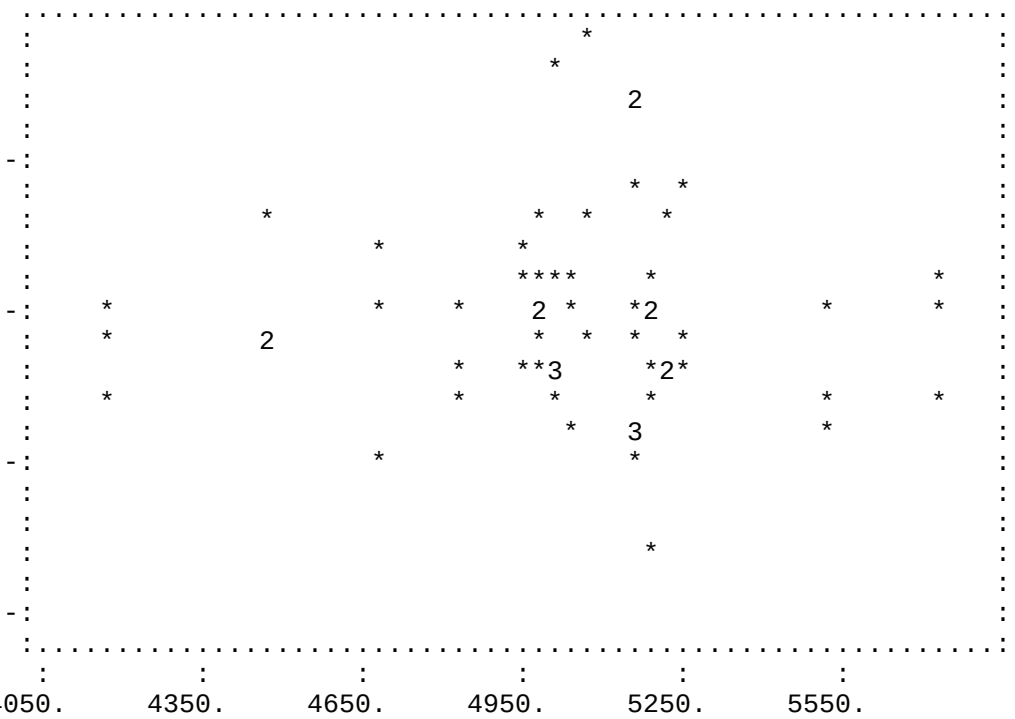
Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	2310.	238.6	9.681	0.3620E-21
2	1	CN$	325.1	180.1	1.805	0.7114E-01
3	1	CN$	-73.82	168.0	-0.4395	0.6603
4	1	CN$	-27.82	130.8	-0.2127	0.8315
5	1	CN$	-375.7	169.7	-2.214	0.2683E-01
6	1	CN$	170.7	156.9	1.088	0.2767
7	1	CN$	210.2	154.6	1.360	0.1740
8	1	CN$	276.3	162.0	1.706	0.8801E-01
9	1	CN$	278.2	134.6	2.068	0.3867E-01
10	1	CN$	535.7	147.9	3.622	0.2918E-03
11	1	CN$	168.8	162.2	1.041	0.2981
12	1	CN$	372.7	167.8	2.222	0.2631E-01
13	1	CN$	459.6	162.9	2.822	0.4778E-02
14	1	CN$	467.1	175.6	2.661	0.7796E-02
15	1	CN$	500.9	166.8	3.003	0.2673E-02
16	1	CN$	512.9	160.5	3.195	0.1397E-02
17	1	CN$	521.3	180.6	2.887	0.3895E-02
18	1	CN$	-411.8	167.4	-2.460	0.1390E-01
19	1	CN$	-512.1	147.1	-3.481	0.4994E-03
20	1	CN$	-156.4	152.7	-1.024	0.3058

(B) The 2007 seasons trials

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/11  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  31
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO PI 2007

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    60

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          60.   3467.       6819.       4957.       731.9

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   15 CODES:(Number  Label) for Variable: ROW
(  1           1)(  2           2)(  3           3)(  4           4)(  5           5)
(  6           6)(  7           7)(  8           8)(  9           9)( 10          10)
( 11          11)( 12          12)( 13          13)( 14          14)( 15          15)

    4 CODES:(Number  Label) for Variable: COLUMN
(  1           1)(  2           2)(  3           3)(  4           4)(

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA         )(  2 CC         )(  3 CD         )(  4 CE         )(  5 D1         )
(  6 D2         )(  7 D3         )(  8 D4         )(  9 EA         )( 10 EB         )
( 11 G1         )( 12 G2         )( 13 H1         )( 14 H2         )( 15 H3         )
( 16 H4         )( 17 H5         )( 18 H6         )( 19 Y2         )( 20 YC         )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  32
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO PI 2007
 Number of non-missing dependent observations:          60
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -279.892035127989061
 Likelihood value -2LogL:      633.299152895978068

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.7936      0.9504E-01   8.351      0.6774E-16
  1 AR1(COLUMN)(2  0.5463      0.1414       3.864      0.1114E-03
```

```
The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.6852E+06  0.3114E+06   2.200      0.2780E-01
Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.903E-02   0.241E-02   0.243E+05
       2   1 AR1(COLUMN)(   0.241E-02   0.200E-01   0.190E+05
       3 Dep(1).........   0.243E+05   0.190E+05   0.970E+11
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  40.00        3.587       0.3297E-03

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	5175.	426.2
1	CN$ CC	4469.	429.8
1	CN$ CD	5171.	437.3
1	CN$ CE	4992.	432.9
1	CN$ D1	5191.	428.8
1	CN$ D2	4990.	433.6
1	CN$ D3	5058.	431.7
1	CN$ D4	4826.	436.9
1	CN$ EA	4670.	422.8
1	CN$ EB	4160.	421.3
1	CN$ G1	5194.	426.7
1	CN$ G2	5733.	426.6
1	CN$ H1	5237.	423.9
1	CN$ H2	5519.	425.2
1	CN$ H3	5029.	429.5
1	CN$ H4	5006.	427.8
1	CN$ H5	4995.	422.7
1	CN$ H6	5147.	434.1
1	CN$ Y2	4947.	422.9
1	CN$ YC	5217.	428.3

Standard Errors of Differences

Minimum	Mean	Maximum
226.8	300.3	362.3

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

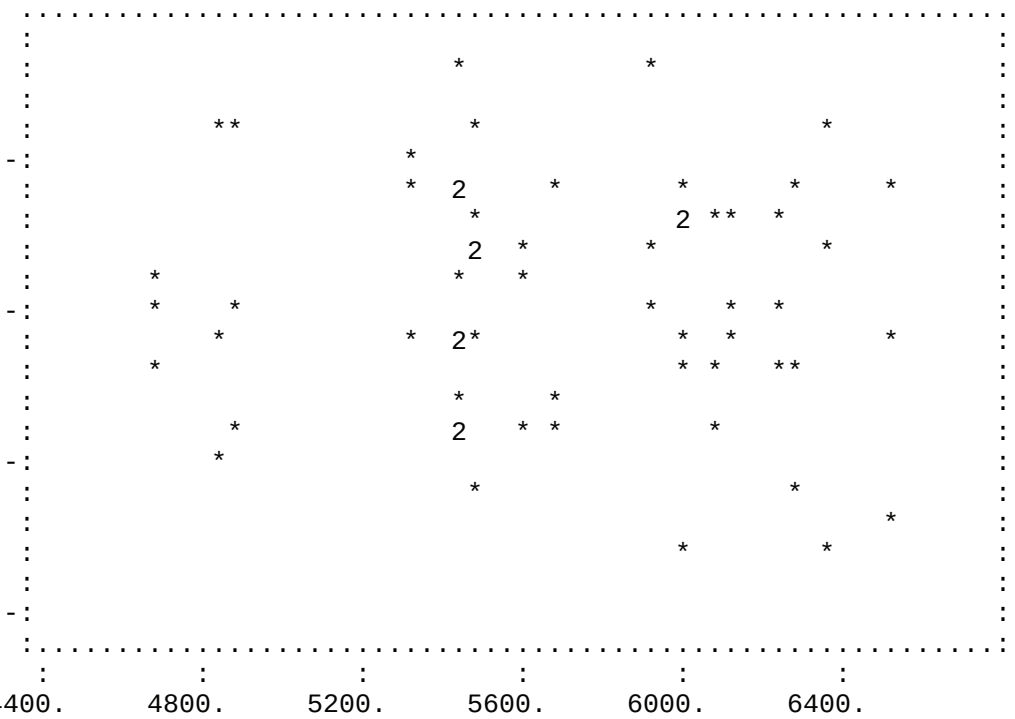
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	5217.	428.3	12.18	0.3969E-33
2	1	CN$	-41.61	288.2	-0.1444	0.8852
3	1	CN$	-747.6	327.5	-2.283	0.2245E-01
4	1	CN$	-45.89	346.3	-0.1325	0.8946
5	1	CN$	-224.2	316.6	-0.7081	0.4789
6	1	CN$	-25.39	318.8	-0.7964E-01	0.9365
7	1	CN$	-226.2	290.1	-0.7797	0.4356
8	1	CN$	-158.2	315.9	-0.5010	0.6164
9	1	CN$	-390.6	275.2	-1.419	0.1558
10	1	CN$	-546.4	296.2	-1.844	0.6511E-01
11	1	CN$	-1057.	305.3	-3.461	0.5378E-03
12	1	CN$	-22.66	304.8	-0.7434E-01	0.9407
13	1	CN$	516.0	298.9	1.726	0.8429E-01
14	1	CN$	20.86	312.8	0.6670E-01	0.9468
15	1	CN$	302.0	299.0	1.010	0.3125
16	1	CN$	-188.0	309.7	-0.6070	0.5438
17	1	CN$	-210.5	300.9	-0.6995	0.4842
18	1	CN$	-221.1	299.9	-0.7374	0.4609
19	1	CN$	-70.00	290.8	-0.2407	0.8098
20	1	CN$	-269.9	276.8	-0.9751	0.3295

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/12  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  34
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO LA 2007

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    60

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          60.   4094.       7276.       5731.       776.8

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   12 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)(

    5 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CC          )(  3 CD          )(  4 CE          )(  5 D1          )
(  6 D2          )(  7 D3          )(  8 D4          )(  9 EA          )( 10 EB          )
( 11 G1          )( 12 G2          )( 13 H1          )( 14 H2          )( 15 H3          )
( 16 H4          )( 17 H5          )( 18 H6          )( 19 Y2          )( 20 YC          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  35
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO LA 2007
 Number of non-missing dependent observations:          60
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -288.748773089021881
 Likelihood value -2LogL:      651.012628818043709

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.6207      0.1407       4.410      0.1034E-04
  1 AR1(COLUMN)(2  0.4551      0.2093       2.175      0.2965E-01
```

```
The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.6578E+06  0.2672E+06   2.461      0.1385E-01
Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.198E-01   0.182E-01   0.294E+05
       2   1 AR1(COLUMN)(   0.182E-01   0.438E-01   0.400E+05
       3 Dep(1).........   0.294E+05   0.400E+05   0.714E+11
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  40.00        4.065       0.9280E-04

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	5997.	397.5
1	CN$ CC	5447.	388.7
1	CN$ CD	4676.	406.2
1	CN$ CE	4864.	402.1
1	CN$ D1	6257.	402.0
1	CN$ D2	5931.	392.2
1	CN$ D3	5431.	389.5
1	CN$ D4	5667.	401.7
1	CN$ EA	6502.	444.7
1	CN$ EB	5325.	421.7
1	CN$ G1	6342.	409.6
1	CN$ G2	6107.	404.9
1	CN$ H1	6279.	390.7
1	CN$ H2	5497.	403.9
1	CN$ H3	6064.	412.2
1	CN$ H4	5584.	395.3
1	CN$ H5	4829.	404.7
1	CN$ H6	5434.	392.1
1	CN$ Y2	5999.	391.3
1	CN$ YC	5476.	401.1

Standard Errors of Differences

Minimum	Mean	Maximum
326.4	429.1	512.6

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment met
hod

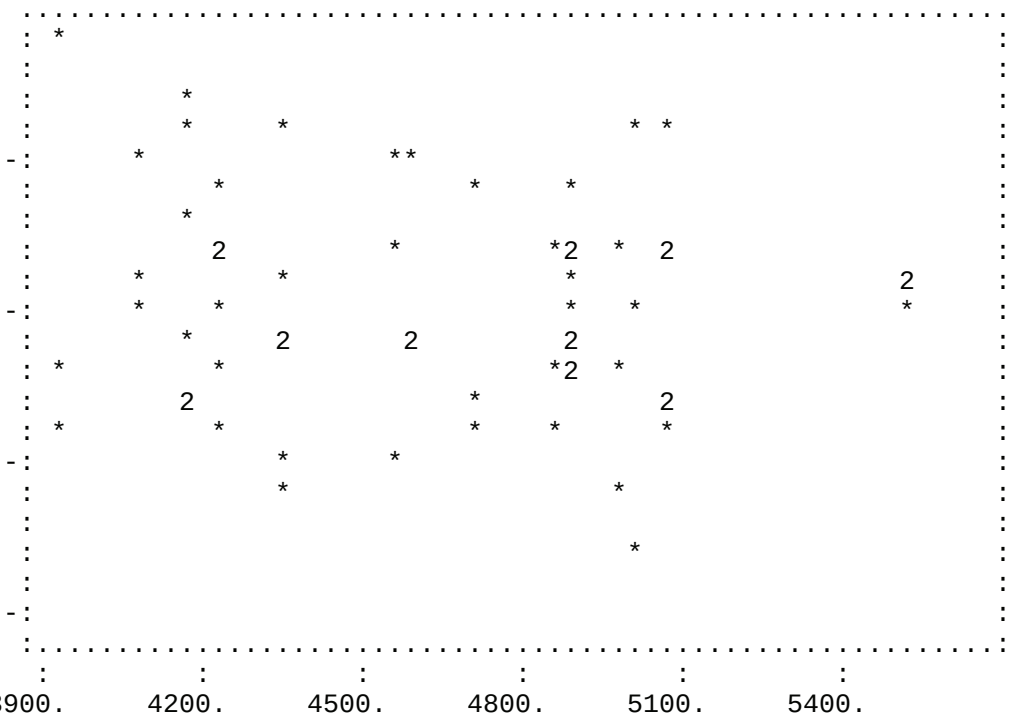
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	5476.	401.1	13.65	0.1900E-41
2	1	CN$	520.3	451.9	1.151	0.2495
3	1	CN$	-29.46	432.3	-0.6814E-01	0.9457
4	1	CN$	-800.8	425.5	-1.882	0.5983E-01
5	1	CN$	-612.4	433.2	-1.413	0.1575
6	1	CN$	780.8	426.7	1.830	0.6725E-01
7	1	CN$	454.8	435.2	1.045	0.2960
8	1	CN$	-45.18	437.6	-0.1032	0.9178
9	1	CN$	190.2	396.7	0.4794	0.6316
10	1	CN$	1026.	481.6	2.130	0.3317E-01
11	1	CN$	-151.5	475.0	-0.3190	0.7498
12	1	CN$	865.1	462.9	1.869	0.6166E-01
13	1	CN$	630.5	432.3	1.458	0.1447
14	1	CN$	802.3	410.1	1.956	0.5041E-01
15	1	CN$	20.90	414.9	0.5037E-01	0.9598
16	1	CN$	587.7	370.2	1.588	0.1124
17	1	CN$	108.0	381.4	0.2831	0.7771
18	1	CN$	-647.5	451.1	-1.435	0.1512
19	1	CN$	-42.67	443.6	-0.9619E-01	0.9234
20	1	CN$	522.2	400.1	1.305	0.1918

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/13  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  37
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO KL 2007

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    60

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          60.   2460.       6735.       4670.       1122.

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   12 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)(

    5 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CC          )(  3 CD          )(  4 CE          )(  5 D1          )
(  6 D2          )(  7 D3          )(  8 D4          )(  9 EA          )( 10 EB          )
( 11 G1          )( 12 G2          )( 13 H1          )( 14 H2          )( 15 H3          )
( 16 H4          )( 17 H5          )( 18 H6          )( 19 Y2          )( 20 YC          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  38
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO KL 2007
 Number of non-missing dependent observations:          60
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -299.490945636931542
 Likelihood value -2LogL:      672.496973913863030

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.5637      0.1427       3.951      0.7790E-04
  1 AR1(COLUMN)(2  0.7422      0.9570E-01   7.756      0.8778E-14
```

```
The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.1546E+07  0.5835E+06   2.649      0.8071E-02
Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.204E-01   0.782E-03   0.447E+05
       2   1 AR1(COLUMN)(   0.782E-03   0.916E-02   0.344E+05
       3 Dep(1).........   0.447E+05   0.344E+05   0.340E+12
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  40.00        1.398       0.1829

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	4898.	582.1
1	CN$ CC	4169.	587.0
1	CN$ CD	5508.	608.9
1	CN$ CE	4718.	587.3
1	CN$ D1	4867.	586.8
1	CN$ D2	5058.	593.5
1	CN$ D3	4159.	605.4
1	CN$ D4	5065.	592.7
1	CN$ EA	4579.	597.7
1	CN$ EB	4240.	578.0
1	CN$ G1	4234.	604.7
1	CN$ G2	3925.	581.6
1	CN$ H1	4339.	590.4
1	CN$ H2	4973.	606.3
1	CN$ H3	4903.	603.5
1	CN$ H4	4905.	612.6
1	CN$ H5	4081.	619.6
1	CN$ H6	4358.	586.0
1	CN$ Y2	4570.	598.1
1	CN$ YC	5009.	597.6

Standard Errors of Differences

Minimum	Mean	Maximum
377.3	491.1	584.6

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

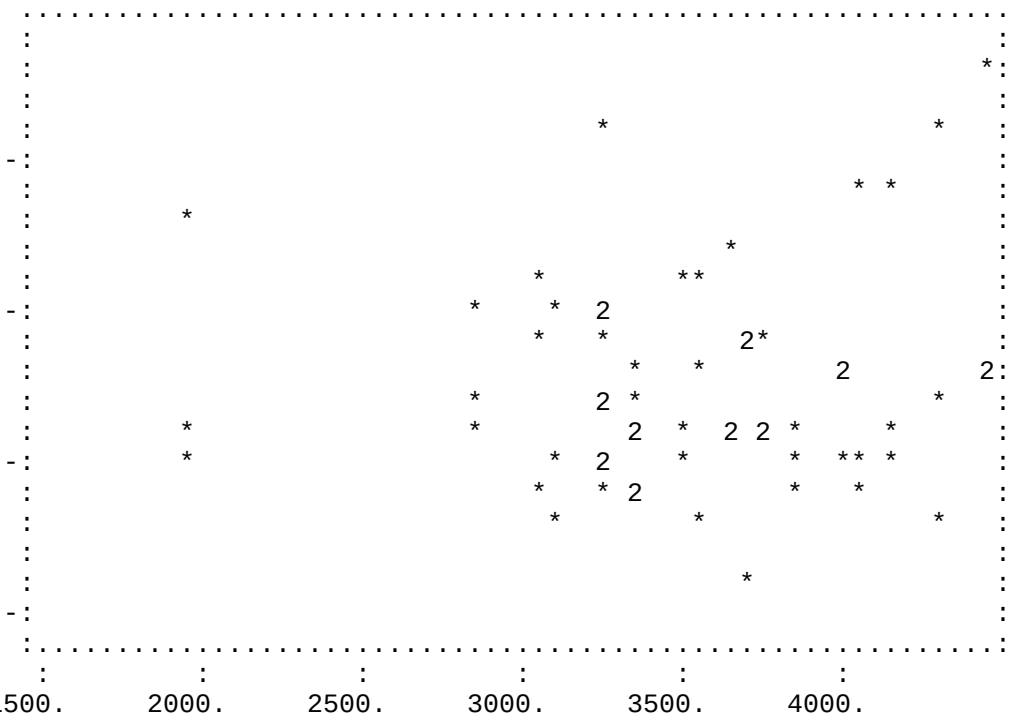
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	5009.	597.6	8.383	0.5160E-16
2	1	CN$	-111.5	438.0	-0.2546	0.7990
3	1	CN$	-840.4	501.6	-1.676	0.9383E-01
4	1	CN$	498.7	499.6	0.9983	0.3181
5	1	CN$	-291.2	490.3	-0.5939	0.5526
6	1	CN$	-142.9	483.7	-0.2954	0.7677
7	1	CN$	48.64	510.8	0.9522E-01	0.9241
8	1	CN$	-850.9	534.2	-1.593	0.1112
9	1	CN$	55.19	432.2	0.1277	0.8984
10	1	CN$	-430.8	497.3	-0.8663	0.3863
11	1	CN$	-769.1	451.7	-1.703	0.8864E-01
12	1	CN$	-775.1	516.0	-1.502	0.1331
13	1	CN$	-1084.	461.5	-2.349	0.1882E-01
14	1	CN$	-670.1	497.0	-1.348	0.1776
15	1	CN$	-36.55	485.4	-0.7529E-01	0.9400
16	1	CN$	-106.6	535.6	-0.1991	0.8422
17	1	CN$	-104.9	576.9	-0.1819	0.8557
18	1	CN$	-928.5	581.0	-1.598	0.1101
19	1	CN$	-651.6	456.0	-1.429	0.1530
20	1	CN$	-439.9	493.0	-0.8922	0.3723

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/14  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  40
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO ME 2007

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    60

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          60.   1515.       6574.       3513.       923.8

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   12 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)(

    5 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CC          )(  3 CD          )(  4 CE          )(  5 D1          )
(  6 D2          )(  7 D3          )(  8 D4          )(  9 EA          )( 10 EB          )
( 11 G1          )( 12 G2          )( 13 H1          )( 14 H2          )( 15 H3          )
( 16 H4          )( 17 H5          )( 18 H6          )( 19 Y2          )( 20 YC          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  41
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO ME 2007
 Number of non-missing dependent observations:          60
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -288.198930163966281
 Likelihood value -2LogL:      649.912942967932509

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.6565      0.1030       6.375      0.1829E-09
  1 AR1(COLUMN)(2  0.5137E-01  0.2397      0.2143      0.8303
```

```
The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.5557E+06  0.1562E+06   3.559      0.3725E-03
Asymptotic Covariance Matrix of the Gamma Estimates
                            1           2           3
       1   1 AR1(ROW)(1).   0.106E-01  -0.398E-02   0.100E+05
       2   1 AR1(COLUMN)(  -0.398E-02   0.575E-01  -0.120E+04
       3 Dep(1).........   0.100E+05  -0.120E+04   0.244E+11
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  40.00        3.527       0.3890E-03

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	3350.	344.6
1	CN$ CC	3687.	350.0
1	CN$ CD	1948.	356.3
1	CN$ CE	3837.	345.3
1	CN$ D1	4433.	344.2
1	CN$ D2	4137.	338.9
1	CN$ D3	3492.	353.3
1	CN$ D4	3526.	338.7
1	CN$ EA	4297.	345.2
1	CN$ EB	3262.	355.9
1	CN$ G1	3344.	338.4
1	CN$ G2	3759.	344.3
1	CN$ H1	3628.	344.8
1	CN$ H2	3238.	344.8
1	CN$ H3	4057.	335.3
1	CN$ H4	2846.	336.6
1	CN$ H5	3071.	339.5
1	CN$ H6	3234.	344.6
1	CN$ Y2	4004.	335.8
1	CN$ YC	3087.	352.9

Standard Errors of Differences

Minimum	Mean	Maximum
345.4	413.5	466.8

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

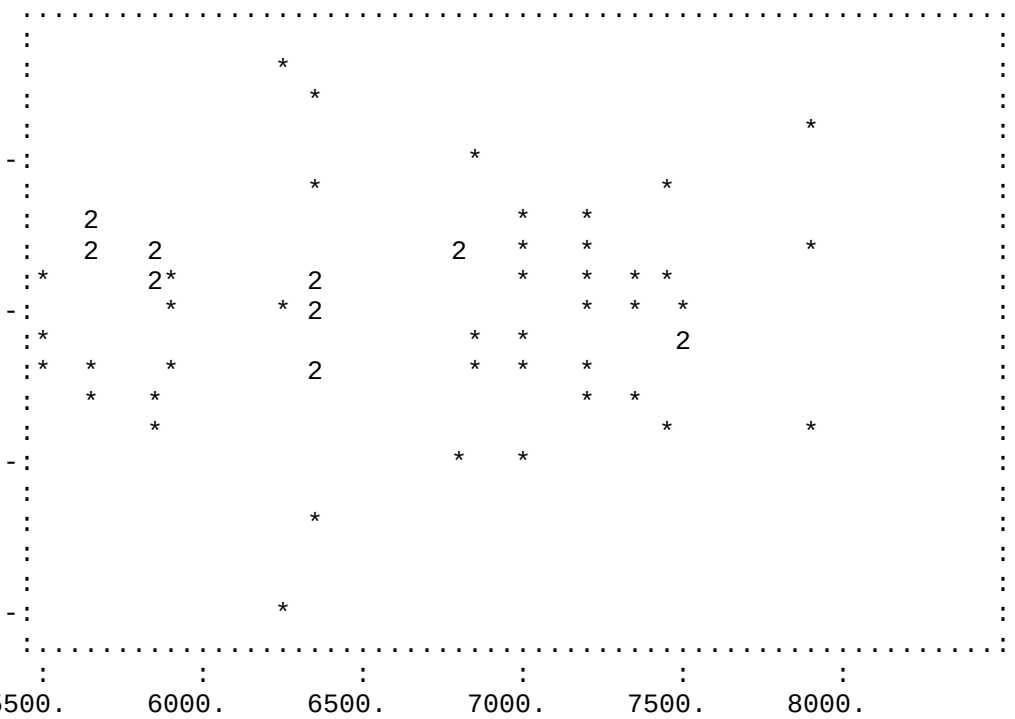
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	3087.	352.9	8.747	0.2186E-17
2	1	CN$	263.0	418.2	0.6288	0.5295
3	1	CN$	600.2	407.9	1.471	0.1412
4	1	CN$	-1139.	463.0	-2.460	0.1388E-01
5	1	CN$	750.4	444.3	1.689	0.9123E-01
6	1	CN$	1346.	431.4	3.120	0.1806E-02
7	1	CN$	1050.	437.0	2.403	0.1628E-01
8	1	CN$	405.0	451.7	0.8967	0.3699
9	1	CN$	439.7	427.7	1.028	0.3039
10	1	CN$	1211.	426.0	2.842	0.4482E-02
11	1	CN$	174.8	357.0	0.4897	0.6243
12	1	CN$	257.3	440.0	0.5848	0.5587
13	1	CN$	672.7	438.0	1.536	0.1246
14	1	CN$	541.5	446.1	1.214	0.2249
15	1	CN$	151.2	439.6	0.3439	0.7310
16	1	CN$	970.5	395.3	2.455	0.1409E-01
17	1	CN$	-240.2	433.7	-0.5540	0.5796
18	1	CN$	-15.71	380.7	-0.4125E-01	0.9671
19	1	CN$	147.2	426.8	0.3448	0.7302
20	1	CN$	917.1	423.0	2.168	0.3017E-01

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/15  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  43
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO RO 2007

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    60

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          60.   4167.       9067.       6590.       958.7

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   12 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)(

    5 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CC          )(  3 CD          )(  4 CE          )(  5 D1          )
(  6 D2          )(  7 D3          )(  8 D4          )(  9 EA          )( 10 EB          )
( 11 G1          )( 12 G2          )( 13 H1          )( 14 H2          )( 15 H3          )
( 16 H4          )( 17 H5          )( 18 H6          )( 19 Y2          )( 20 YC          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  44
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO RO 2007
 Number of non-missing dependent observations:          60
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -295.671073651448182
 Likelihood value -2LogL:      664.857229942896311

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.1422      0.1794      0.7925      0.4281
  1 AR1(COLUMN)(2  0.1994      0.2450      0.8139      0.4157
```

```
The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.5709E+06  0.1320E+06   4.327      0.1513E-04
Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.322E-01  -0.102E-02   0.328E+04
       2   1 AR1(COLUMN)(  -0.102E-02   0.600E-01   0.781E+04
       3 Dep(1).........   0.328E+04   0.781E+04   0.174E+11
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  40.00        3.011       0.1646E-02

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	7221.	425.4
1	CN$ CC	6980.	426.2
1	CN$ CD	6327.	424.7
1	CN$ CE	5869.	425.1
1	CN$ D1	6820.	422.0
1	CN$ D2	6979.	424.7
1	CN$ D3	7363.	423.9
1	CN$ D4	7493.	424.1
1	CN$ EA	5889.	425.1
1	CN$ EB	5513.	423.6
1	CN$ G1	7447.	423.3
1	CN$ G2	7925.	425.9
1	CN$ H1	5827.	425.7
1	CN$ H2	6326.	428.1
1	CN$ H3	6368.	426.3
1	CN$ H4	5663.	427.6
1	CN$ H5	6262.	426.0
1	CN$ H6	6875.	426.4
1	CN$ Y2	5649.	422.3
1	CN$ YC	7200.	424.5

Standard Errors of Differences

Minimum	Mean	Maximum
545.9	587.2	601.5

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

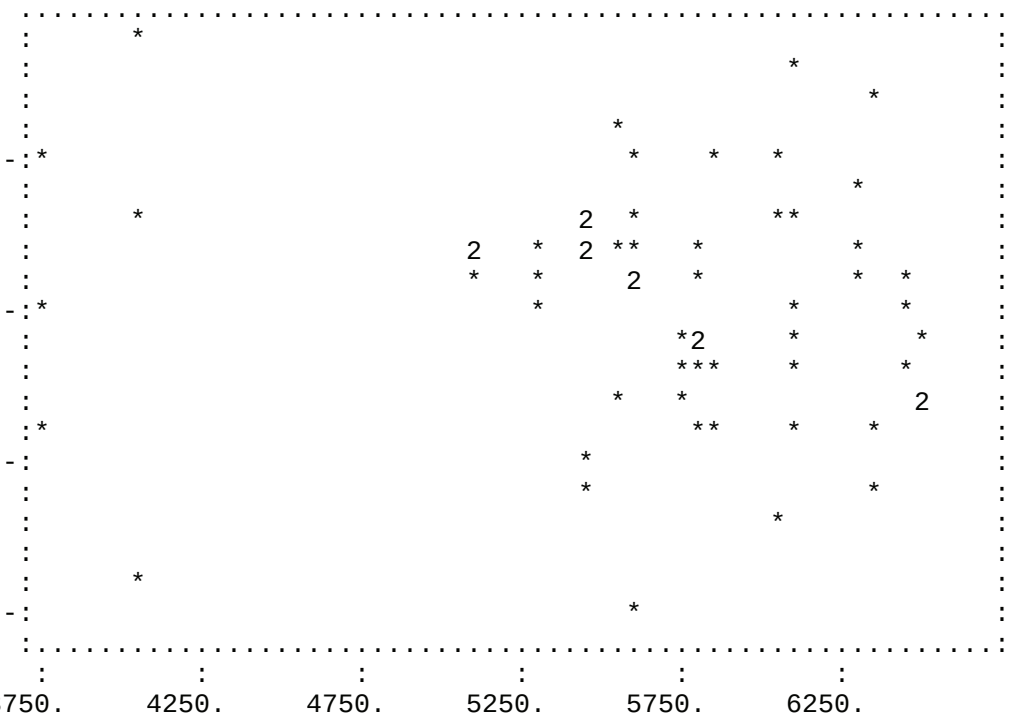
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	7200.	424.5	16.96	0.1638E-63
2	1	CN$	21.06	594.5	0.3542E-01	0.9717
3	1	CN$	-219.4	596.5	-0.3677	0.7131
4	1	CN$	-873.2	597.5	-1.461	0.1439
5	1	CN$	-1331.	594.2	-2.240	0.2512E-01
6	1	CN$	-379.9	567.3	-0.6696	0.5031
7	1	CN$	-221.0	595.0	-0.3714	0.7103
8	1	CN$	163.6	597.1	0.2740	0.7841
9	1	CN$	292.9	577.1	0.5076	0.6118
10	1	CN$	-1310.	595.1	-2.202	0.2767E-01
11	1	CN$	-1686.	593.1	-2.843	0.4468E-02
12	1	CN$	247.7	579.8	0.4271	0.6693
13	1	CN$	725.1	578.8	1.253	0.2103
14	1	CN$	-1372.	593.8	-2.311	0.2084E-01
15	1	CN$	-873.8	558.9	-1.563	0.1179
16	1	CN$	-832.1	593.0	-1.403	0.1606
17	1	CN$	-1536.	578.0	-2.658	0.7868E-02
18	1	CN$	-938.2	595.1	-1.577	0.1149
19	1	CN$	-324.8	578.0	-0.5619	0.5742
20	1	CN$	-1551.	581.3	-2.668	0.7633E-02

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/16  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  46
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO TY 2007

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    60

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          60.   2607.       7409.       5665.       890.2

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   12 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)(

    5 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CC          )(  3 CD          )(  4 CE          )(  5 D1          )
(  6 D2          )(  7 D3          )(  8 D4          )(  9 EA          )( 10 EB          )
( 11 G1          )( 12 G2          )( 13 H1          )( 14 H2          )( 15 H3          )
( 16 H4          )( 17 H5          )( 18 H6          )( 19 Y2          )( 20 YC          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  47
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO TY 2007
 Number of non-missing dependent observations:          60
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -290.152381440915406
 Likelihood value -2LogL:      653.819845521830757

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.4023      0.1640       2.454      0.1414E-01
  1 AR1(COLUMN)(2  0.2804      0.1789       1.567      0.1171
```

```
The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.5158E+06  0.1327E+06   3.886      0.1018E-03
Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.269E-01  -0.494E-02   0.880E+04
       2   1 AR1(COLUMN)(  -0.494E-02   0.320E-01   0.449E+04
       3 Dep(1).........   0.880E+04   0.449E+04   0.176E+11
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  40.00        4.196       0.6628E-04

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	5587.	380.2
1	CN$ CC	5600.	375.8
1	CN$ CD	3728.	371.5
1	CN$ CE	5323.	375.0
1	CN$ D1	6091.	375.3
1	CN$ D2	5546.	386.0
1	CN$ D3	5838.	386.5
1	CN$ D4	5808.	375.8
1	CN$ EA	6041.	373.0
1	CN$ EB	5114.	371.7
1	CN$ G1	5754.	418.5
1	CN$ G2	6349.	373.4
1	CN$ H1	5461.	375.7
1	CN$ H2	5452.	378.4
1	CN$ H3	6445.	380.7
1	CN$ H4	5796.	382.7
1	CN$ H5	4063.	375.9
1	CN$ H6	6509.	380.9
1	CN$ Y2	6078.	367.2
1	CN$ YC	6280.	369.1

Standard Errors of Differences

Minimum	Mean	Maximum
428.1	492.6	549.0

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

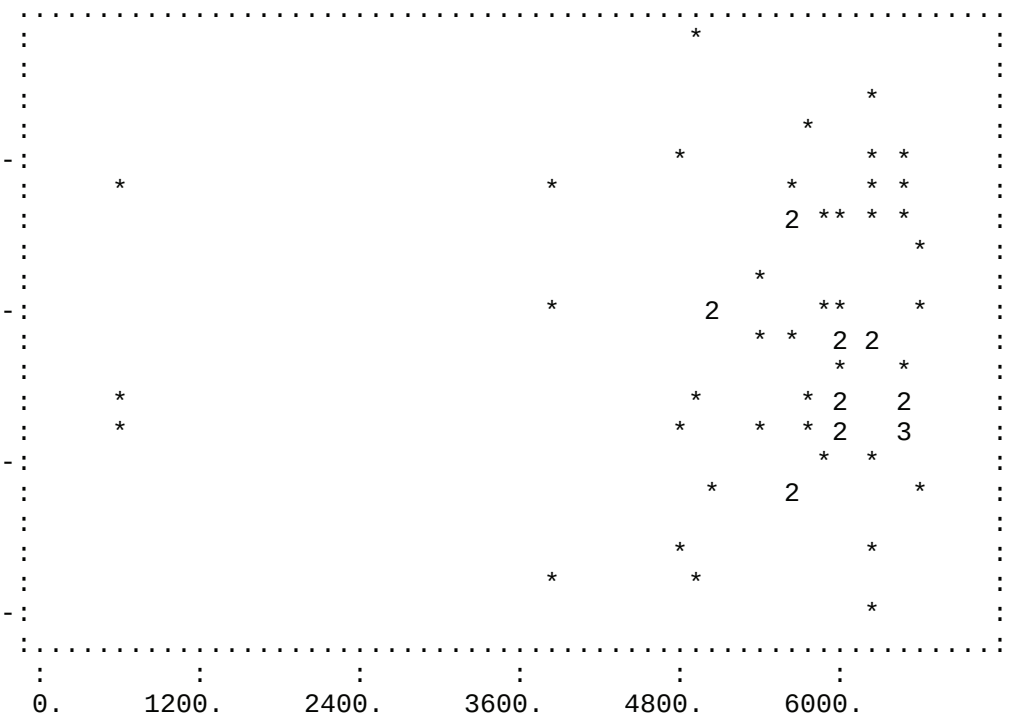
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	6280.	369.1	17.01	0.6454E-64
2	1	CN$	-692.6	505.7	-1.370	0.1708
3	1	CN$	-680.1	491.9	-1.383	0.1668
4	1	CN$	-2552.	493.1	-5.175	0.2280E-06
5	1	CN$	-956.8	478.7	-1.999	0.4563E-01
6	1	CN$	-188.9	495.2	-0.3815	0.7028
7	1	CN$	-733.6	498.8	-1.471	0.1414
8	1	CN$	-441.6	482.0	-0.9161	0.3596
9	1	CN$	-472.1	465.2	-1.015	0.3102
10	1	CN$	-239.5	489.6	-0.4891	0.6248
11	1	CN$	-1166.	474.6	-2.458	0.1398E-01
12	1	CN$	-525.7	516.7	-1.017	0.3089
13	1	CN$	68.88	428.1	0.1609	0.8722
14	1	CN$	-819.1	497.9	-1.645	0.9992E-01
15	1	CN$	-827.6	474.2	-1.745	0.8095E-01
16	1	CN$	164.5	447.2	0.3679	0.7130
17	1	CN$	-484.1	502.3	-0.9639	0.3351
18	1	CN$	-2217.	496.9	-4.462	0.8117E-05
19	1	CN$	229.0	496.4	0.4613	0.6446
20	1	CN$	-201.9	465.1	-0.4342	0.6642

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/17  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  49
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO NA 2007

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    60

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          60.   284.2       7262.       5525.       1419.

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   15 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)( 13           13)( 14           14)( 15           15)

    4 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CC          )(  3 CD          )(  4 CE          )(  5 D1          )
(  6 D2          )(  7 D3          )(  8 D4          )(  9 EA          )( 10 EB          )
( 11 G1          )( 12 G2          )( 13 H1          )( 14 H2          )( 15 H3          )
( 16 H4          )( 17 H5          )( 18 H6          )( 19 Y2          )( 20 YC          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  50
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO NA 2007
 Number of non-missing dependent observations:          60
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -287.755649830645780
 Likelihood value -2LogL:      649.026382301291505

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.1081      0.1846      0.5855      0.5582
  1 AR1(COLUMN)(2 -0.1377      0.2294     -0.6005      0.5482
```

```
The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.3826E+06  0.8684E+05   4.406      0.1051E-04
Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.341E-01   0.195E-02   0.183E+04
       2   1 AR1(COLUMN)(   0.195E-02   0.526E-01  -0.261E+04
       3 Dep(1).........   0.183E+04  -0.261E+04   0.754E+10
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  40.00        14.65       0.1846E-11

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	6180.	352.4
1	CN$ CC	6043.	350.2
1	CN$ CD	649.0	352.1
1	CN$ CE	5069.	350.8
1	CN$ D1	5813.	350.8
1	CN$ D2	6020.	351.5
1	CN$ D3	6198.	350.5
1	CN$ D4	6196.	350.7
1	CN$ EA	5388.	350.5
1	CN$ EB	4741.	349.4
1	CN$ G1	5890.	351.7
1	CN$ G2	6003.	351.2
1	CN$ H1	6596.	352.2
1	CN$ H2	6479.	351.8
1	CN$ H3	5615.	350.8
1	CN$ H4	4899.	362.7
1	CN$ H5	3874.	350.6
1	CN$ H6	6462.	351.4
1	CN$ Y2	6469.	351.8
1	CN$ YC	5610.	351.7

Standard Errors of Differences

Minimum	Mean	Maximum
476.8	497.1	518.8

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	5610.	351.7	15.95	0.2858E-56
2	1	CN$	570.1	496.9	1.147	0.2513
3	1	CN$	433.0	495.1	0.8744	0.3819
4	1	CN$	-4961.	497.4	-9.973	0.1998E-22
5	1	CN$	-540.8	497.0	-1.088	0.2765
6	1	CN$	203.4	507.3	0.4009	0.6885
7	1	CN$	410.2	507.5	0.8083	0.4189
8	1	CN$	588.0	506.0	1.162	0.2452
9	1	CN$	586.0	486.7	1.204	0.2286
10	1	CN$	-222.2	496.0	-0.4480	0.6542
11	1	CN$	-869.0	487.8	-1.781	0.7485E-01
12	1	CN$	279.6	487.5	0.5734	0.5663
13	1	CN$	393.2	496.3	0.7922	0.4283
14	1	CN$	985.8	498.5	1.978	0.4796E-01
15	1	CN$	869.2	479.3	1.813	0.6976E-01
16	1	CN$	5.317	495.7	0.1073E-01	0.9914
17	1	CN$	-711.2	507.5	-1.401	0.1611
18	1	CN$	-1736.	487.5	-3.561	0.3699E-03
19	1	CN$	852.3	496.1	1.718	0.8580E-01
20	1	CN$	859.4	509.4	1.687	0.9157E-01

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/18  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  52
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO RI 2007

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    60

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          60.   1278.       8069.       5273.       1336.

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   15 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)( 13           13)( 14           14)( 15           15)

    4 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CC          )(  3 CD          )(  4 CE          )(  5 D1          )
(  6 D2          )(  7 D3          )(  8 D4          )(  9 EA          )( 10 EB          )
( 11 G1          )( 12 G2          )( 13 H1          )( 14 H2          )( 15 H3          )
( 16 H4          )( 17 H5          )( 18 H6          )( 19 Y2          )( 20 YC          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  53
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO RI 2007
 Number of non-missing dependent observations:          60
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -307.274038246624798
 Likelihood value -2LogL:      688.063159133249542

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.2996      0.1440       2.081      0.3745E-01
  1 AR1(COLUMN)(2  0.2908      0.2216       1.312      0.1894
```

The scale parameters

Dep.	Sigma_Squared	Std. Error	Z	Pr > |Z|
Dep(1)	0.1113E+07	0.2743E+06	4.056	0.4988E-04

Asymptotic Covariance Matrix of the Gamma Estimates

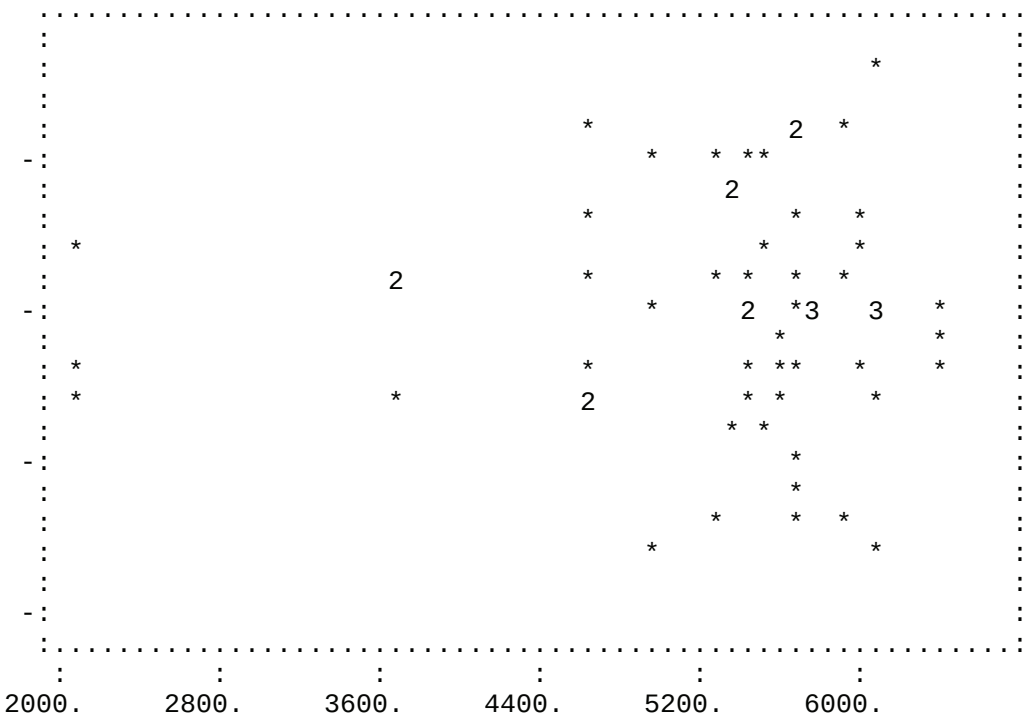
		1	2	3
1	1 AR1(ROW)(1).	0.207E-01	0.170E-02	0.120E+05
2	1 AR1(COLUMN)(	0.170E-02	0.491E-01	0.200E+05
3	Dep(1).....	0.120E+05	0.200E+05	0.752E+11

Warning: Denominator degrees of freedom estimates do not account for measurement error parameters.

ANOVA Table for Sequentially Deleted Fixed Effects
Denominator Degrees of Freedom: Containment method

Dep	Effect	DFNum	DFDen	F - Statistic	P > |F|
1	CN$	19	40.00	3.173	0.1038E-02

Plot of residuals against predicted values



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	3690.	566.8
1	CN$ CC	5675.	611.9
1	CN$ CD	2069.	572.2
1	CN$ CE	6068.	623.3
1	CN$ D1	5287.	567.0
1	CN$ D2	6379.	574.2
1	CN$ D3	5711.	587.0
1	CN$ D4	5973.	562.0
1	CN$ EA	5471.	560.0
1	CN$ EB	4946.	564.1
1	CN$ G1	5622.	568.6
1	CN$ G2	5791.	565.3
1	CN$ H1	5670.	573.1
1	CN$ H2	5556.	568.7
1	CN$ H3	5444.	559.2
1	CN$ H4	6075.	578.2
1	CN$ H5	4632.	562.1
1	CN$ H6	5348.	569.4
1	CN$ Y2	4661.	578.9
1	CN$ YC	5886.	566.6

Standard Errors of Differences

Minimum	Mean	Maximum
671.0	767.4	833.2

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

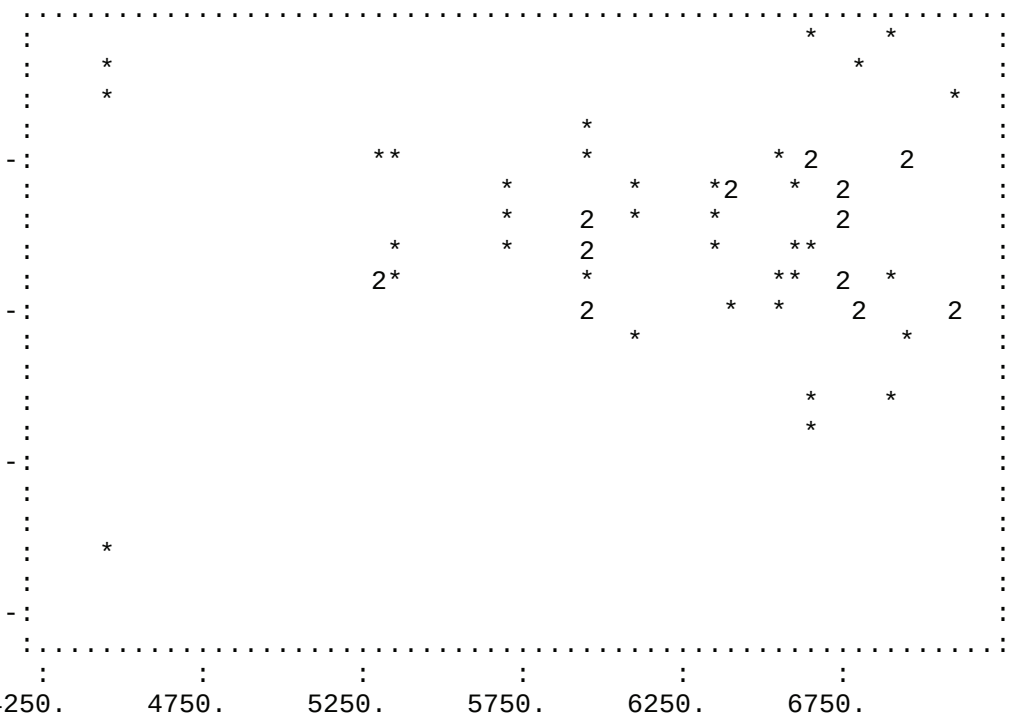
The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	5886.	566.6	10.39	0.2773E-24
2	1	CN$	-2196.	774.3	-2.836	0.4562E-02
3	1	CN$	-211.0	812.4	-0.2597	0.7951
4	1	CN$	-3817.	746.0	-5.116	0.3116E-06
5	1	CN$	182.0	823.3	0.2211	0.8250
6	1	CN$	-599.1	779.1	-0.7689	0.4419
7	1	CN$	492.4	783.7	0.6284	0.5298
8	1	CN$	-175.5	796.6	-0.2203	0.8257
9	1	CN$	87.10	741.6	0.1175	0.9065
10	1	CN$	-415.6	728.7	-0.5704	0.5684
11	1	CN$	-940.1	770.3	-1.220	0.2223
12	1	CN$	-264.6	775.0	-0.3413	0.7328
13	1	CN$	-95.04	731.2	-0.1300	0.8966
14	1	CN$	-216.3	770.4	-0.2808	0.7789
15	1	CN$	-330.3	746.7	-0.4424	0.6582
16	1	CN$	-442.5	774.3	-0.5714	0.5677
17	1	CN$	188.4	746.3	0.2524	0.8007
18	1	CN$	-1254.	721.0	-1.740	0.8191E-01
19	1	CN$	-538.2	746.4	-0.7211	0.4708
20	1	CN$	-1226.	749.4	-1.635	0.1020

```
IRREML 2.0.7: REML ANALYSIS FOR COMMAND SET 0607_SP/19  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------------ :PAGE  55
 THE IRREML PROGRAM WAS WRITTEN BY DOUGLAS CLARKSON OF SCIENCEOPS FOR IRRI
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1
 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO AL 2007

 Command File: C:\!MSC THESIS\! LIT THESIS WRITING FOLDER\CROPSTAT DATA\2006\060
 7_SP.GFC   Data File: COMB_CS

 Number of Records:    60

 Variables in Data Set: ROW  COLUMN  CN$  GY_AS

 SUMMARY STATISTICS FOR NUMERIC VARIATES
 VARIATE       NOBS   MINIMUM     MAXIMUM     MEAN     STD. DEV.
 GY_AS          60.   2744.       7973.       6255.       831.7

 Classification Variables: ROW  COLUMN  CN$

 Levels of the classification variables

   12 CODES:(Number  Label) for Variable: ROW
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)
(  6            6)(  7            7)(  8            8)(  9            9)( 10           10)
( 11           11)( 12           12)(

    5 CODES:(Number  Label) for Variable: COLUMN
(  1            1)(  2            2)(  3            3)(  4            4)(  5            5)

   20 CODES:(Number  Label) for Variable: CN$
(  1 CA          )(  2 CC          )(  3 CD          )(  4 CE          )(  5 D1          )
(  6 D2          )(  7 D3          )(  8 D4          )(  9 EA          )( 10 EB          )
( 11 G1          )( 12 G2          )( 13 H1          )( 14 H2          )( 15 H3          )
( 16 H4          )( 17 H5          )( 18 H6          )( 19 Y2          )( 20 YC          )
```

```
IRREML: REML ANALYSIS FOR VARIATE GY_AS  FILE COMB_CS    3/ 7/ 9 13: 4
 ------------------------------------------------------------------ :PAGE  56
                          SPATIAL ANALYSIS OF TRITICALE FIELD TRIALS
                          DATA FOR GRAIN YIELD (AS-IS), KG.HA-1

 DATA RECORDS SELECTED FROM FILE COMB_CS
 INCLUDE RECORDS WITH     SITE$   (  3) EQUAL TO AL 2007
 Number of non-missing dependent observations:          60
 Check estimability of effect means: T

 Model Specification
 Intercept in model: Yes
 The Fixed Effects Model
     GY_AS = Intercept + CN$
 The Random Effects Terms
 None

 RANDOM EFFECT COVARIANCE MODEL.  0 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 None

 RESIDUAL EFFECT COVARIANCE MODEL.  1 SPECIFIED STRUCTURES
 TERM              PARAMETER INDICES  STRUCTURE SCALE SAME NBLOCK GROUPING VARS
 ------------------------------------------------------------------------------
 RESIDUAL                      1-  2   product   0     1      1
   AR1(ROW)                    1-  1       AR1   0     1      1
   AR1(COLUMN)                 2-  2       AR1   0     1      1

 Number of columns in the fixed effects model:     20
 Number of columns in the random effects model:     0
Message: Relative function convergence

 Final REML criterion:        -287.479421098724004
 Likelihood value -2LogL:      648.473924837447953

 Variance/Covariance component parameters
 Dep Name           Gamma Coef. Std. Error    Z       Pr > |Z|    Scaled Gamma  Std. Error
  Product
  1 AR1(ROW)(1)    0.1091      0.2131      0.5121      0.6086
  1 AR1(COLUMN)(2 -0.9860E-01  0.2010     -0.4906      0.6237
```

```
The scale parameters
 Dep.  Sigma_Squared Std. Error      Z         Pr > |Z|
Dep(1) .......    0.3758E+06  0.8537E+05   4.401      0.1076E-04
Asymptotic Covariance Matrix of the Gamma Estimates
                              1           2           3
       1   1 AR1(ROW)(1).   0.454E-01  -0.661E-02   0.293E+04
       2   1 AR1(COLUMN)(  -0.661E-02   0.404E-01  -0.193E+04
       3 Dep(1).........   0.293E+04  -0.193E+04   0.729E+10
Warning: Denominator degrees of freedom estimates do not account
 for measurement error parameters.

       ANOVA Table for Sequentially Deleted Fixed Effects
       Denominator Degrees of Freedom: Containment method
 Dep Effect    DFNum   DFDen   F - Statistic   P > |F|
  1      CN$     19  40.00        3.827       0.1734E-03

 Plot of residuals against predicted values
```



Balanced Least Squares Means Fixed

Dep	Level	LSMean	Std. Error
1	CN$ CA	5291.	350.5
1	CN$ CC	6913.	350.0
1	CN$ CD	4449.	350.0
1	CN$ CE	5945.	349.7
1	CN$ D1	6340.	349.2
1	CN$ D2	6670.	349.6
1	CN$ D3	6085.	349.2
1	CN$ D4	6774.	350.9
1	CN$ EA	6769.	349.8
1	CN$ EB	5700.	349.4
1	CN$ G1	6782.	349.5
1	CN$ G2	7091.	348.7
1	CN$ H1	6948.	349.2
1	CN$ H2	6397.	348.9
1	CN$ H3	6554.	350.6
1	CN$ H4	5944.	349.7
1	CN$ H5	5345.	348.8
1	CN$ H6	5956.	361.6
1	CN$ Y2	6592.	349.6
1	CN$ YC	6667.	349.2

Standard Errors of Differences

Minimum	Mean	Maximum
475.0	494.4	511.2

Denominator Degrees of Freedom in linear combinations: Residual Method
Denominator Degrees of Freedom in fixed effect tests and means: Containment method

The Regression Coefficient Estimates

Num	Dep	Name	Estimate	Std. Error	Z	Pr > |Z|
1	1	Intercept	6667.	349.2	19.09	0.3002E-80
2	1	CN$	-1376.	492.7	-2.793	0.5230E-02
3	1	CN$	246.1	494.5	0.4977	0.6187
4	1	CN$	-2218.	494.9	-4.481	0.7430E-05
5	1	CN$	-722.2	501.7	-1.439	0.1500
6	1	CN$	-327.3	494.4	-0.6621	0.5079
7	1	CN$	2.577	501.2	0.5141E-02	0.9959
8	1	CN$	-582.2	493.6	-1.180	0.2382
9	1	CN$	107.1	502.5	0.2130	0.8313
10	1	CN$	102.2	501.4	0.2037	0.8386
11	1	CN$	-967.3	494.5	-1.956	0.5043E-01
12	1	CN$	114.9	485.6	0.2366	0.8129
13	1	CN$	424.2	492.9	0.8606	0.3894
14	1	CN$	280.5	500.7	0.5602	0.5754
15	1	CN$	-269.8	492.4	-0.5479	0.5838
16	1	CN$	-112.8	494.8	-0.2280	0.8196
17	1	CN$	-722.9	484.8	-1.491	0.1359
18	1	CN$	-1322.	493.0	-2.682	0.7319E-02
19	1	CN$	-711.4	503.2	-1.414	0.1574
20	1	CN$	-74.81	484.9	-0.1543	0.8774

Addendum 4: Residual values of the two-dimensional spatial analysis for grain yield, the 2006 and 2007 season trials

Table A4.4.18. Residual values for grain yield (kg.ha^{-1}) of the Vredenburg 2006 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4
	Rep. 1	Rep. 2	Rep. 3	Rep. 4
1	56.76	462.10	1015.20	313.51
2	-37.06	130.28	519.31	495.84
3	-217.12	292.98	190.59	-100.71
4	-677.72	73.73	579.12	684.98
5	-364.65	172.82	-80.80	452.68
6	-169.39	-166.13	-405.28	425.29
7	-359.64	-218.00	-18.39	561.49
8	-152.91	88.38	-62.54	203.40
9	-146.06	589.92	-308.74	-48.00
10	439.51	1004.09	66.60	-611.53
11	377.25	828.25	634.58	-1267.68
12	689.48	554.67	370.43	-738.00
13	430.16	678.60	238.29	-584.91
14	477.49	-1073.15	-117.16	-117.26
15	664.78	878.52	245.57	-595.40
16	892.37	703.61	1437.61	-569.64
17	878.80	207.67	-297.58	-665.93
18	-813.44	-782.12	-853.84	-914.75
19	-904.02	24.52	-1290.64	-900.10
20	-309.69	-397.16	-770.31	-507.46

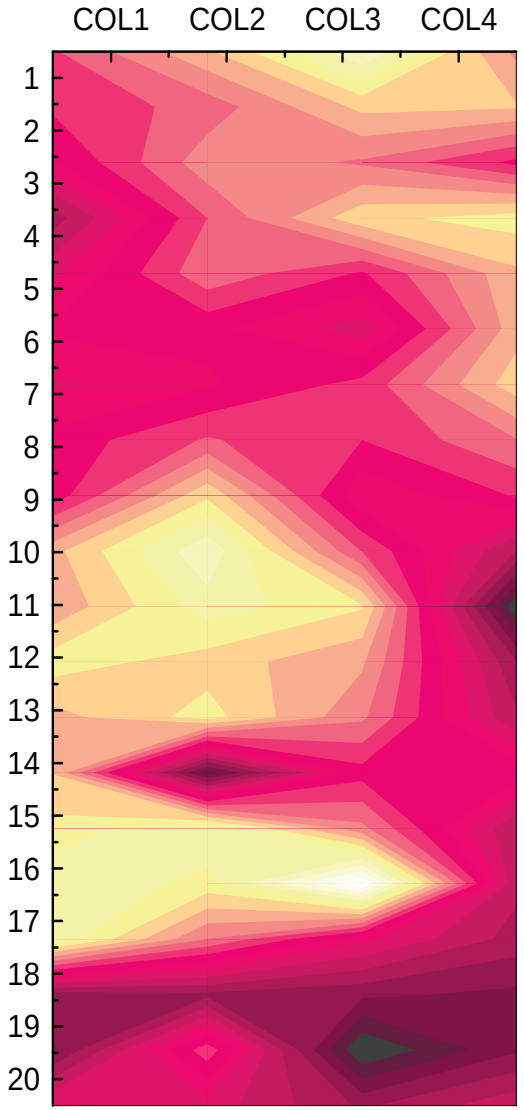


Table A4.4.19. Residual values for grain yield (kg.ha^{-1}) of the Langgewens 2006 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4
	Rep. 1	Rep. 2	Rep. 3	Rep. 4
1	-497.22	-1633.66	788.94	416.52
2	-727.75	-2014.69	606.69	384.87
3	-46.06	-1442.34	1041.76	421.56
4	153.72	-123.32	1381.21	1091.91
5	-402.95	-532.92	294.16	730.49
6	-822.18	-930.38	1283.99	1182.90
7	-908.77	-655.89	1519.62	1885.31
8	-1020.96	-281.19	841.35	1909.55
9	-1963.81	-1153.93	689.32	1210.16
10	-1580.72	-1237.35	138.04	862.62
11	-664.28	-957.42	139.87	-670.21
12	-1590.16	-1425.13	343.20	130.03
13	-944.21	-941.76	928.06	651.34
14	-1291.45	-673.40	1368.08	1172.89
15	-911.61	-1473.87	1167.01	1063.68
16	613.53	-324.55	1309.07	147.98
17	455.43	501.87	1174.71	-166.40
18	-384.16	-735.00	819.33	350.84
19	-475.23	-1270.68	507.86	9.84
20	247.18	-702.97	1485.79	-83.23

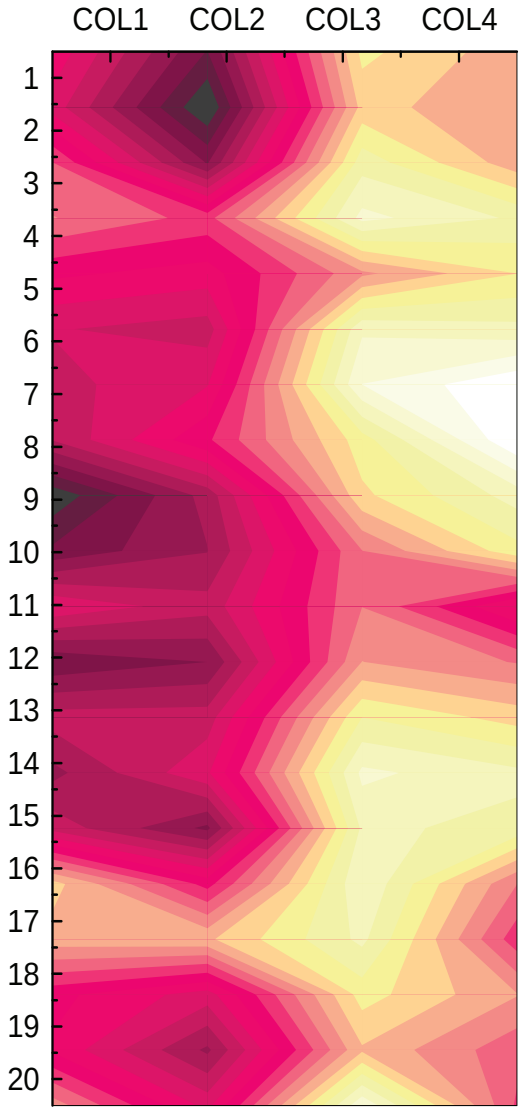


Table A4.4.20. Residual values for grain yield (kg.ha^{-1}) of the Mariendahl 2006 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4
	Rep. 1	Rep. 2	Rep. 3	Rep. 4
1	-183.43	3.49	47.16	64.25
2	-220.03	-9.68	-139.22	-286.67
3	-332.70	-58.03	-292.47	-90.45
4	-159.56	-237.16	-100.09	141.12
5	-108.09	-243.17	-299.45	5.89
6	-694.08	-800.85	-817.31	-177.55
7	-407.67	-222.00	-165.79	-5.11
8	-84.80	9.78	244.49	270.98
9	-196.02	-21.34	-185.04	-187.00
10	-548.68	-235.26	-224.90	198.39
11	-469.61	86.25	-311.42	123.46
12	-441.43	-554.97	-770.52	-239.00
13	-48.23	414.06	624.61	646.17
14	115.51	282.67	339.06	277.09
15	-293.57	75.81	286.89	624.07
16	109.70	813.16	531.93	311.12
17	261.41	490.81	598.59	654.58
18	637.00	667.85	430.91	223.31
19	-222.11	353.96	278.32	623.50
20	-421.18	204.57	171.96	184.00

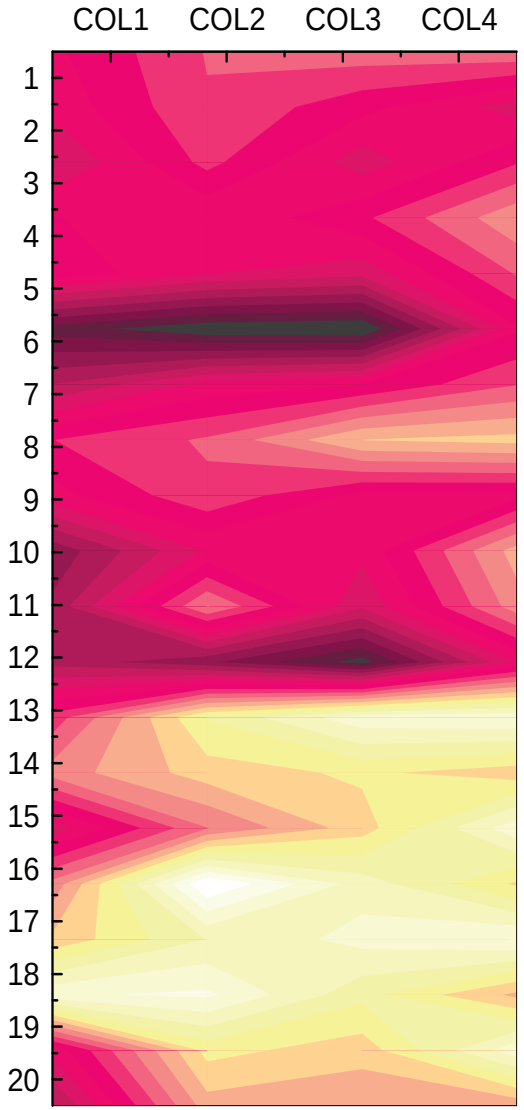


Table A4.4.21. Residual values for grain yield (kg.ha^{-1}) of the Roodebloem 2006 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4
	Rep. 1	Rep. 2	Rep. 3	Rep. 4
1	-9.10	166.34	-238.72	505.33
2	-834.08	-306.72	172.29	768.36
3	-890.10	-377.44	533.19	1133.30
4	-783.60	-93.77	-68.95	344.77
5	-726.85	31.87	341.05	168.87
6	-757.68	28.32	583.83	317.92
7	-554.87	315.61	719.36	590.66
8	-901.57	24.01	909.76	820.16
9	-956.73	-425.67	-194.42	323.02
10	-1634.57	-252.44	-705.86	25.97
11	-617.78	-445.17	-42.28	-885.60
12	-1214.78	-439.03	-981.45	-24.48
13	-316.16	488.36	294.42	544.93
14	674.25	725.23	481.08	492.03
15	311.96	119.37	-85.24	463.92
16	-280.23	-321.02	-410.24	980.51
17	268.17	-52.22	-305.71	338.83
18	-232.75	552.12	-238.42	-542.91
19	-291.04	146.62	63.21	391.74
20	496.39	1068.27	237.47	-310.80

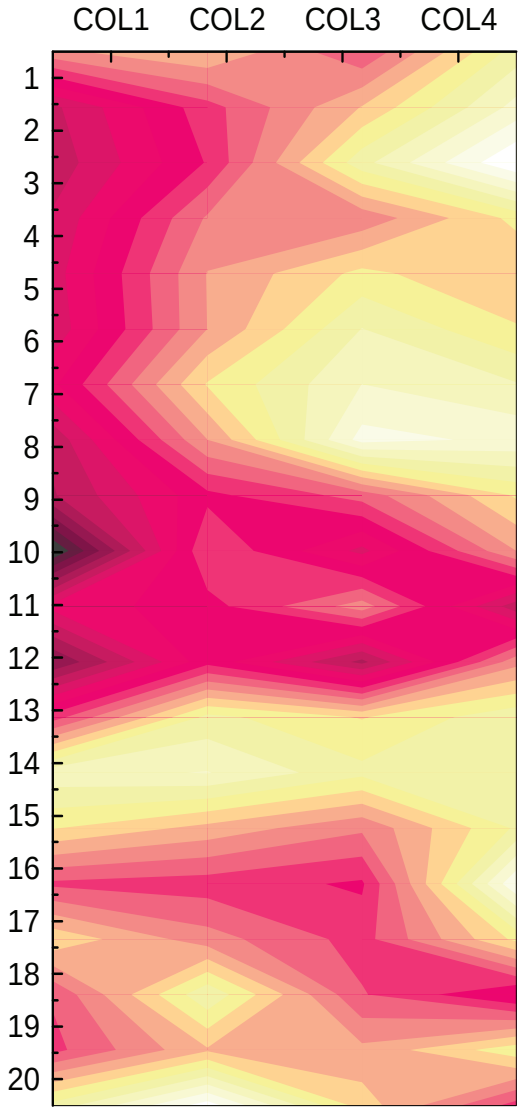


Table A4.4.22. Residual values for grain yield (kg.ha^{-1}) of the Tygerhoek 2006 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4
	Rep. 1	Rep. 2	Rep. 3	Rep. 4
1	517.45	-91.96	94.24	831.10
2	987.75	-29.10	-337.26	277.48
3	605.94	-365.26	191.99	302.69
4	560.42	-439.13	224.06	317.38
5	249.22	-15.82	-3.62	202.60
6	67.67	143.74	-437.47	-109.46
7	549.92	-170.22	-18.57	225.01
8	-105.81	-448.12	-70.53	549.09
9	-204.95	-265.94	-153.58	1010.66
10	21.85	-477.36	-159.90	-42.50
11	145.66	-212.07	94.93	-185.17
12	33.98	-212.61	-321.53	-581.96
13	-86.18	-344.67	-444.04	-417.18
14	456.84	282.25	-1027.88	-377.46
15	762.35	234.06	-227.06	147.88
16	526.10	-409.73	129.65	141.15
17	228.08	-60.70	-120.86	-476.37
18	-427.98	320.60	-474.26	-76.53
19	170.13	-91.27	-391.99	-446.22
20	53.80	-70.53	-220.76	-493.91

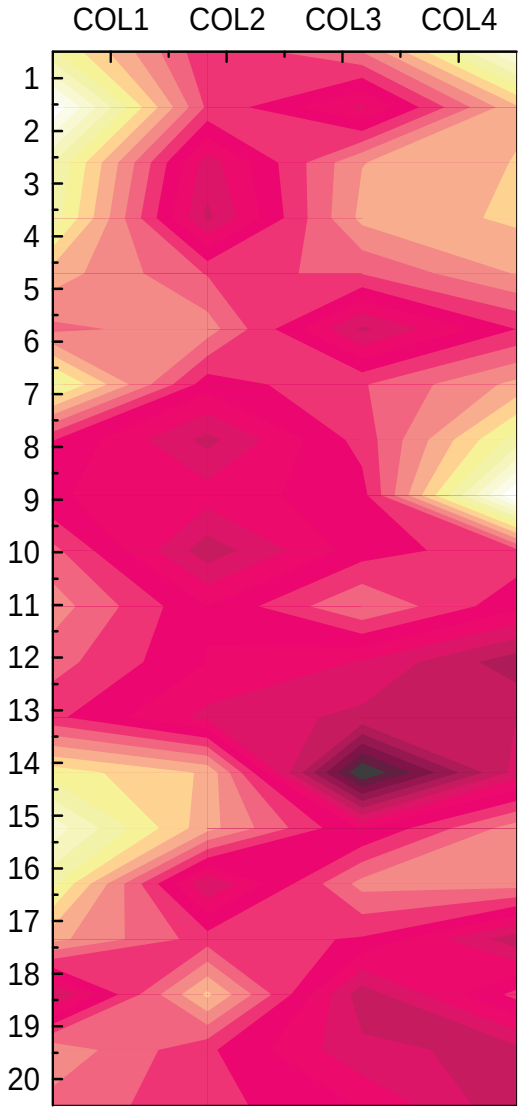


Table A4.4.23. Residual values for grain yield (kg.ha^{-1}) of the Napier 2006 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4
	Rep. 1	Rep. 2	Rep. 3	Rep. 4
1	-436.43	390.28	-473.93	-321.58
2	-452.73	67.29	-445.05	-36.65
3	-572.92	-25.06	-797.01	-577.49
4	-45.95	172.42	-38.78	-348.78
5	-624.89	400.93	0.24	263.29
6	-218.81	-167.85	237.87	619.30
7	233.95	-1383.17	-282.84	-119.60
8	-480.26	217.70	483.63	339.32
9	-568.34	666.46	351.49	-3.21
10	1096.23	1090.47	823.55	795.63
11	-742.42	384.55	802.23	784.42
12	27.83	290.11	192.02	-71.16
13	-495.78	542.32	542.63	834.47
14	-646.94	-356.76	345.06	268.91
15	435.08	848.78	678.30	496.02
16	-96.33	-74.77	-555.55	147.35
17	-724.96	-164.77	213.70	77.53
18	-190.25	182.14	215.63	7.55
19	-443.45	292.39	682.04	382.23
20	-1267.98	-845.25	97.86	-188.10

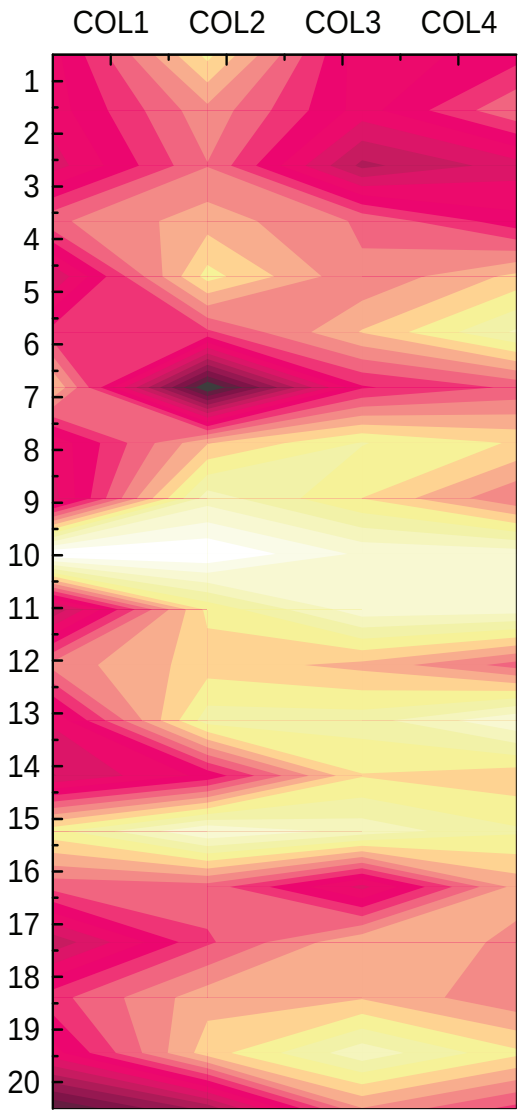


Table A4.4.24. Residual values for grain yield (kg.ha^{-1}) of the Mariendahl 2006 senior block A trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4
	Rep. 1	Rep. 2	Rep. 3	Rep. 4
1	-253.57	-315.30	201.15	244.87
2	-264.73	-543.89	43.89	-317.26
3	-124.50	-662.10	-16.65	-314.91
4	122.29	336.32	-38.04	-431.81
5	-692.68	-379.30	-447.33	-588.97
6	-287.74	-760.95	-155.31	-307.36
7	-267.17	94.85	302.92	181.40
8	175.60	83.80	155.11	134.44
9	142.11	-17.28	2.18	-47.52
10	-215.33	-280.01	99.36	166.98
11	-381.43	-310.48	70.08	257.03
12	-307.36	3.47	-249.72	-318.54
13	84.93	418.83	235.07	456.21
14	325.62	253.92	393.35	315.78
15	241.30	238.39	73.29	327.97
16	20.68	37.11	96.98	38.75
17	199.45	335.38	598.18	132.68
18	-235.80	196.77	611.88	94.85
19	93.60	300.53	625.50	389.24
20	148.51	358.08	98.77	-128.13

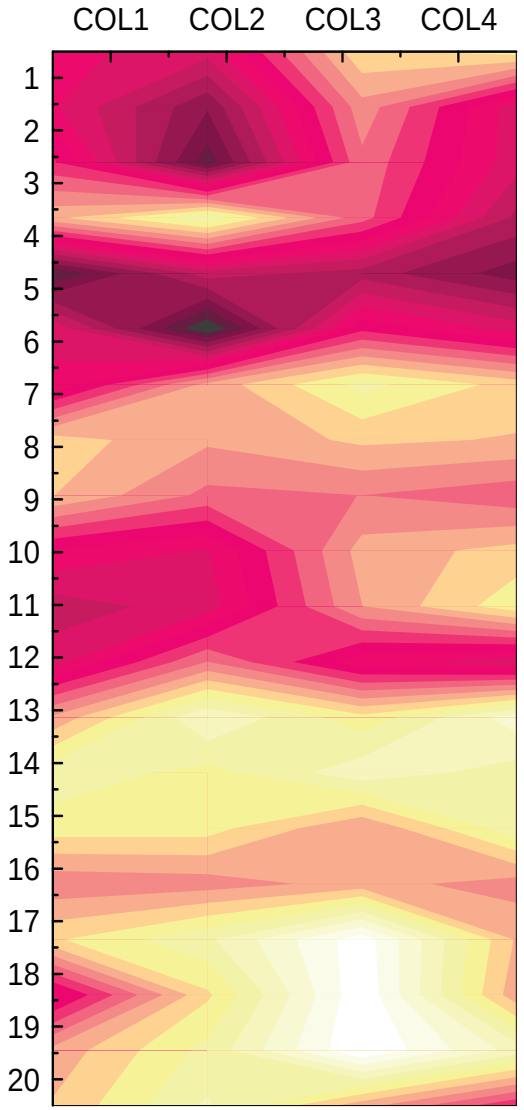


Table A4.4.25. Residual values for grain yield (kg.ha^{-1}) of the Mariendahl 2006 senior block B trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4
	Rep. 1	Rep. 2	Rep. 3	Rep. 4
1	-1074.00	-540.88	-192.00	-117.27
2	-25.15	-292.56	-265.14	-370.36
3	-509.31	-846.62	-465.04	-759.59
4	232.95	-226.26	108.90	-318.64
5	-669.22	-777.83	-695.30	-719.84
6	-864.26	-676.77	-684.61	-386.65
7	-238.28	-61.05	-236.14	112.74
8	-9.34	-335.52	-159.39	-2.82
9	-84.80	-259.25	-309.14	-298.28
10	-31.99	-531.79	-787.25	-276.26
11	307.18	-230.82	-33.68	-171.15
12	-57.37	112.22	16.14	290.37
13	643.77	530.04	311.97	605.07
14	223.52	336.18	453.60	521.90
15	88.74	270.56	-35.57	375.25
16	275.42	135.57	0.29	141.62
17	481.59	792.38	1080.06	1131.59
18	118.27	197.42	391.47	297.38
19	189.76	192.97	578.28	1082.38
20	102.42	433.83	313.31	595.92

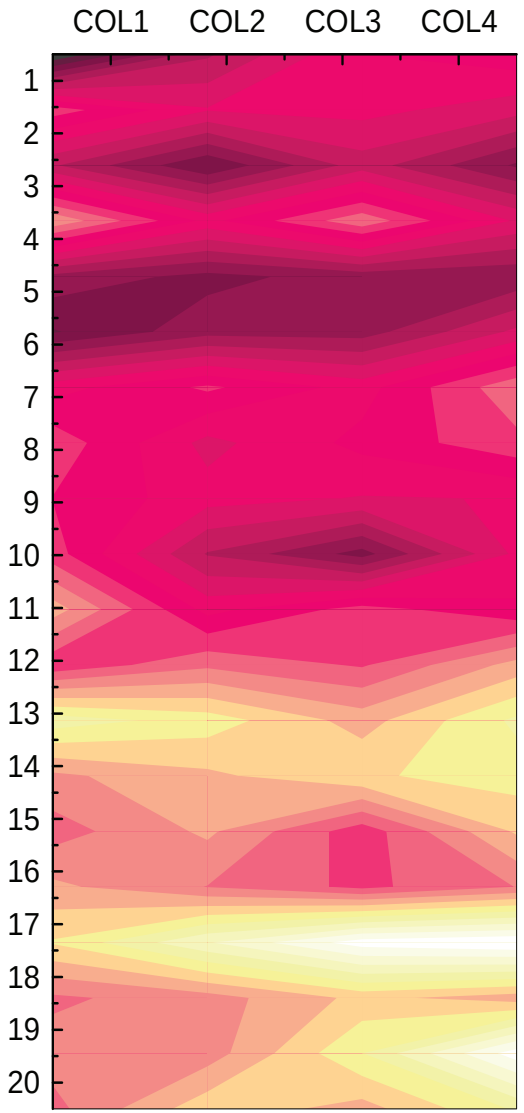


Table A4.4.26. Residual values for grain yield (kg.ha^{-1}) of the Mariendahl 2006 senior block C trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4
	Rep. 1	Rep. 2	Rep. 3	Rep. 4
1	-233.34	-366.22	-249.02	160.49
2	-137.43	124.01	-24.92	-191.78
3	-441.30	-29.15	-50.43	-516.64
4	-17.48	243.89	-6.99	-282.63
5	-210.83	-208.06	-294.38	-855.99
6	-523.07	-587.45	-340.49	-1096.95
7	-688.35	-23.36	42.70	154.42
8	-190.28	-27.18	-126.86	-91.27
9	-389.51	61.41	-418.35	-154.58
10	-254.26	-70.02	-70.19	-313.35
11	-285.60	-229.48	-119.30	-320.90
12	65.33	-259.10	-90.32	-376.62
13	326.16	72.13	-57.07	-338.53
14	794.10	581.70	801.25	569.49
15	355.85	72.42	278.48	32.16
16	58.38	154.61	613.18	123.10
17	555.23	563.61	625.49	165.01
18	445.29	465.08	584.73	463.12
19	752.90	796.56	402.10	-98.16
20	850.26	427.65	-13.56	135.14

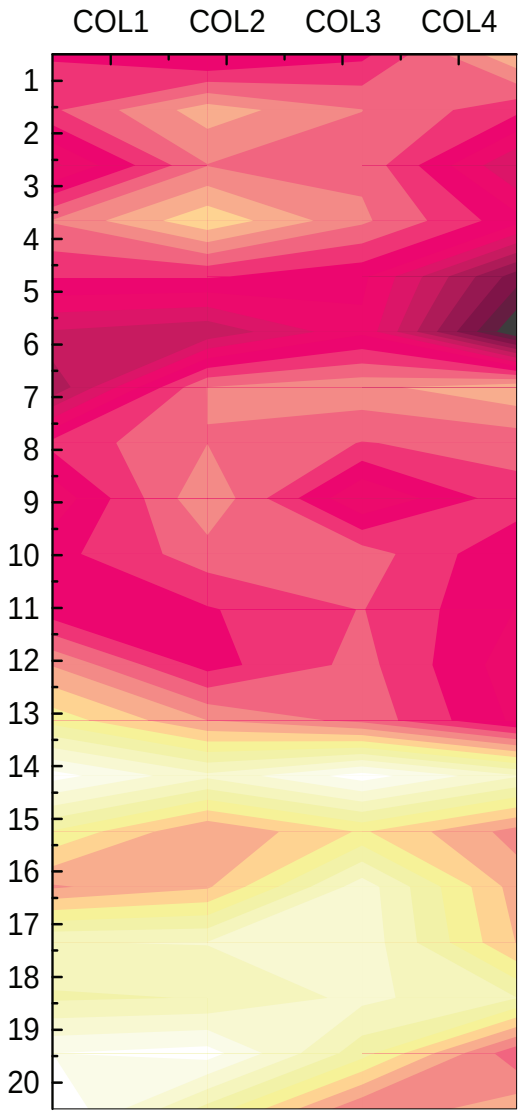


Table A4.4.27. Residual values for grain yield (kg.ha^{-1}) of the Mariendahl 2006 senior block D trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4
	Rep. 1	Rep. 2	Rep. 3	Rep. 4
1	-1043.37	-662.60	-886.57	-679.78
2	-583.74	-417.12	-896.97	-821.36
3	75.87	194.52	-72.16	-103.54
4	-103.51	-267.70	-593.06	-980.89
5	-534.26	-237.73	-430.20	-858.12
6	-493.44	52.19	7.11	-350.81
7	-83.21	-7.47	-157.71	-496.79
8	-59.68	508.26	320.28	490.82
9	-583.72	-41.13	-478.62	-209.10
10	-263.80	500.60	89.77	368.96
11	-5.17	357.80	-28.78	222.28
12	-274.36	-112.68	-49.88	132.71
13	80.70	318.59	78.74	301.11
14	548.68	739.97	151.99	228.94
15	289.70	565.79	10.17	-46.70
16	253.89	717.70	597.23	804.60
17	-147.73	448.46	539.48	691.49
18	895.33	835.67	782.07	1121.83
19	506.30	247.30	-64.80	-4.80
20	798.73	589.57	574.65	693.19

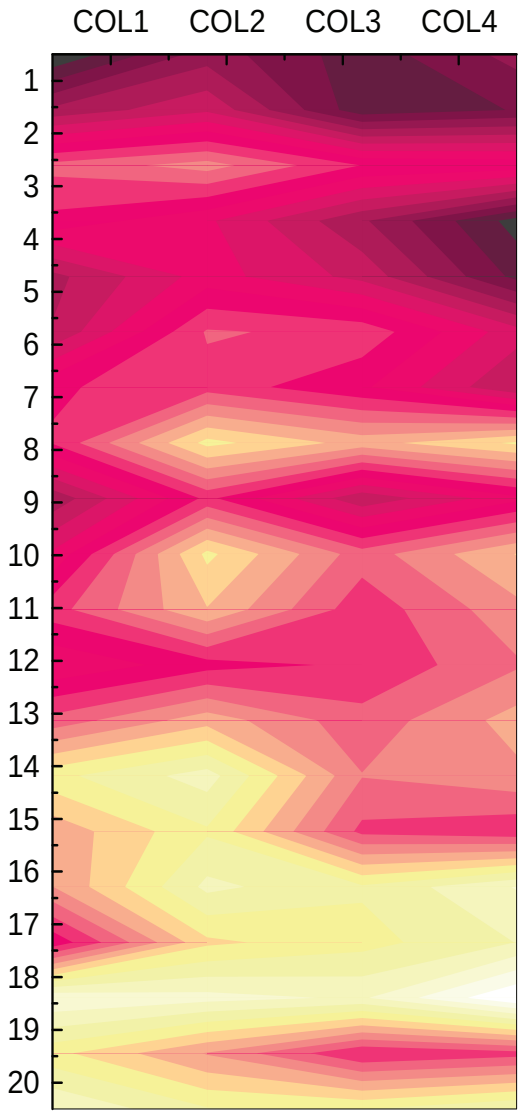


Table A4.4.28. Residual values for grain yield (kg.ha^{-1}) of the Piketberg 2007 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4
	Rep. 1			
1	-849.27	652.51	25.20	-792.07
2	-584.60	566.84	101.68	-528.37
3	-306.80	257.13	392.47	-692.56
4	-887.09	-476.62	-681.63	-1669.86
5	-849.91	-349.11	-518.82	-905.16
	Rep. 2			
6	-4.16	123.93	34.15	-278.60
7	-297.52	90.22	25.25	-282.41
8	-370.96	-1042.25	-231.62	-450.22
9	-304.84	-828.72	-385.75	-449.07
10	848.09	-18.81	-296.64	90.89
	Rep. 3			
11	756.22	-59.48	-486.36	90.63
12	1307.09	-153.05	-687.17	-430.37
13	1481.17	376.69	-162.32	71.01
14	1760.48	605.40	175.02	46.52
15	1661.85	679.08	127.46	232.01

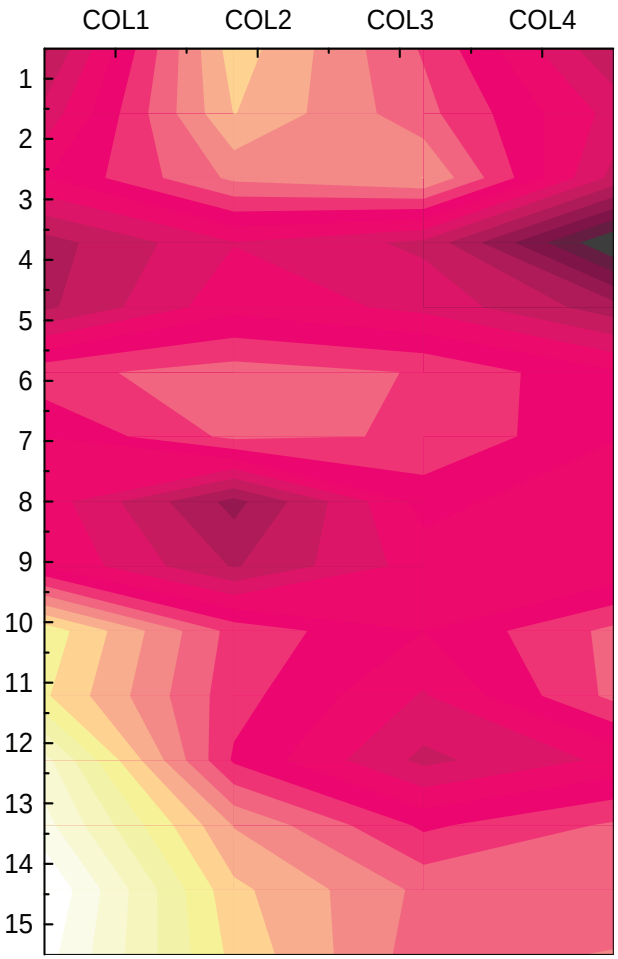


Table A4.4.29. Residual values for grain yield (kg.ha^{-1}) of the Klipheuwel 2007 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4	Column 5
			Rep. 1		
1	-304.62	668.00	-85.91	605.66	1107.55
2	526.66	1852.32	1467.22	1831.20	2200.97
3	1516.83	2697.59	1573.84	1676.49	1165.64
4	197.24	749.94	569.56	739.08	340.47
			Rep. 2		
5	-1684.00	-1170.68	-1472.78	-638.33	-16.26
6	-403.80	-439.76	374.51	1697.69	1159.52
7	-329.16	-1007.42	-538.84	38.63	401.59
8	28.66	-870.35	-528.06	-1036.15	810.04
			Rep. 3		
9	-390.86	-482.52	-280.32	-799.12	740.06
10	-1229.66	-1213.08	-1149.89	-682.88	216.99
11	-1344.54	-1037.40	-666.25	535.77	591.34
12	-2549.63	-1660.84	-1358.16	-354.87	146.79

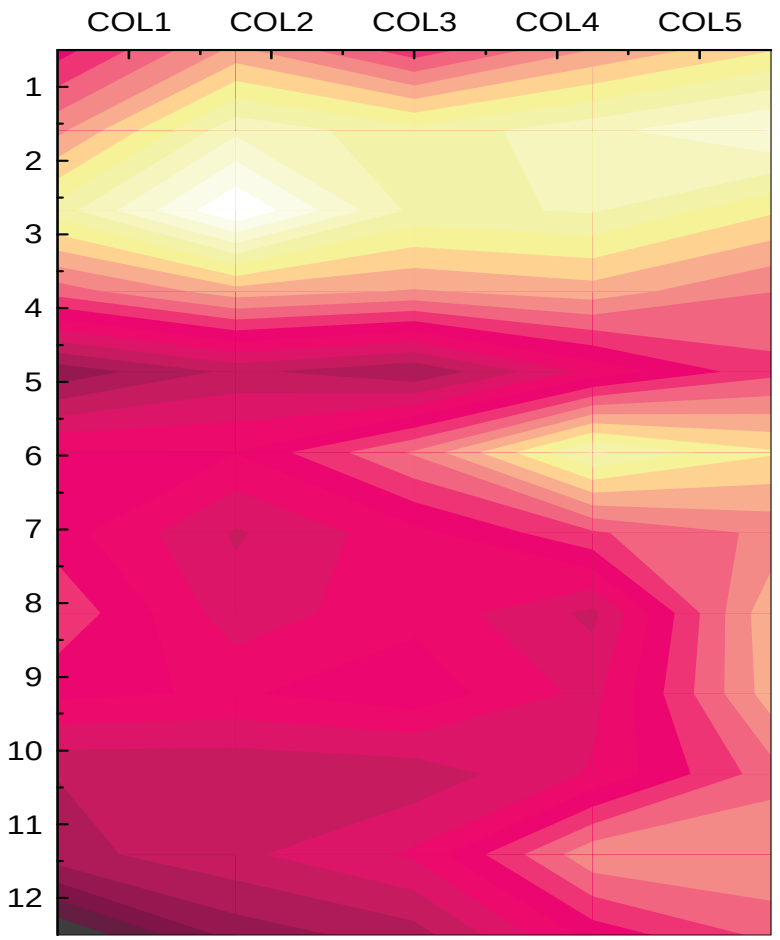


Table A4.4.30. Residual values for grain yield (kg.ha^{-1}) of the Langgewens 2007 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4	Column 5
			Rep. 1		
1	-393.87	333.62	493.68	-551.07	-252.03
2	-627.61	614.56	432.06	-576.38	-192.26
3	-734.59	484.53	653.59	385.77	908.11
4	-516.17	933.97	-187.82	10.76	49.91
			Rep. 2		
5	-1347.47	631.46	-308.72	439.13	-624.61
6	-1329.98	15.33	-251.93	317.28	-387.00
7	-207.35	956.89	144.86	172.07	-663.17
8	-1071.21	-95.23	-658.87	-184.38	-203.42
			Rep. 3		
9	-97.09	354.35	-905.76	31.77	8.47
10	381.25	644.66	-699.63	671.59	577.89
11	768.66	1299.13	549.76	1352.26	902.02
12	592.73	189.07	-900.52	-125.03	535.94

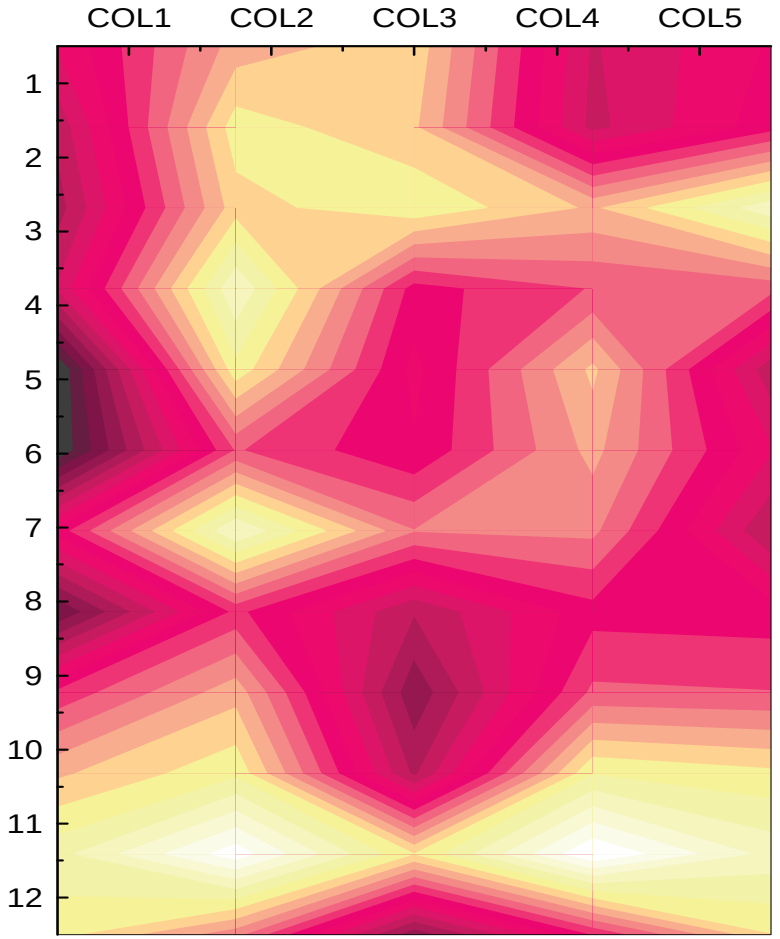


Table A4.4.31. Residual values for grain yield (kg.ha^{-1}) of the Mariendahl 2007 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4	Column 5
			Rep. 1		
1	-145.07	-1295.70	-748.16	-432.44	39.83
2	-514.99	-836.63	-841.18	-424.31	1292.47
3	-939.89	-623.39	-237.41	-333.28	928.83
4	215.72	-768.00	-284.74	-419.43	535.12
			Rep. 2		
5	1.62	-640.95	-205.08	-236.60	2140.95
6	-518.57	-421.51	-247.17	-386.20	1753.15
7	-122.06	-505.39	-121.48	-283.19	466.60
8	221.34	-695.79	-686.25	182.77	266.88
			Rep. 3		
9	98.49	-266.59	-411.04	44.83	186.83
10	650.38	268.44	-282.45	-399.01	1268.13
11	772.56	526.70	-197.84	-128.45	1656.24
12	449.13	761.49	262.50	-452.87	1117.92

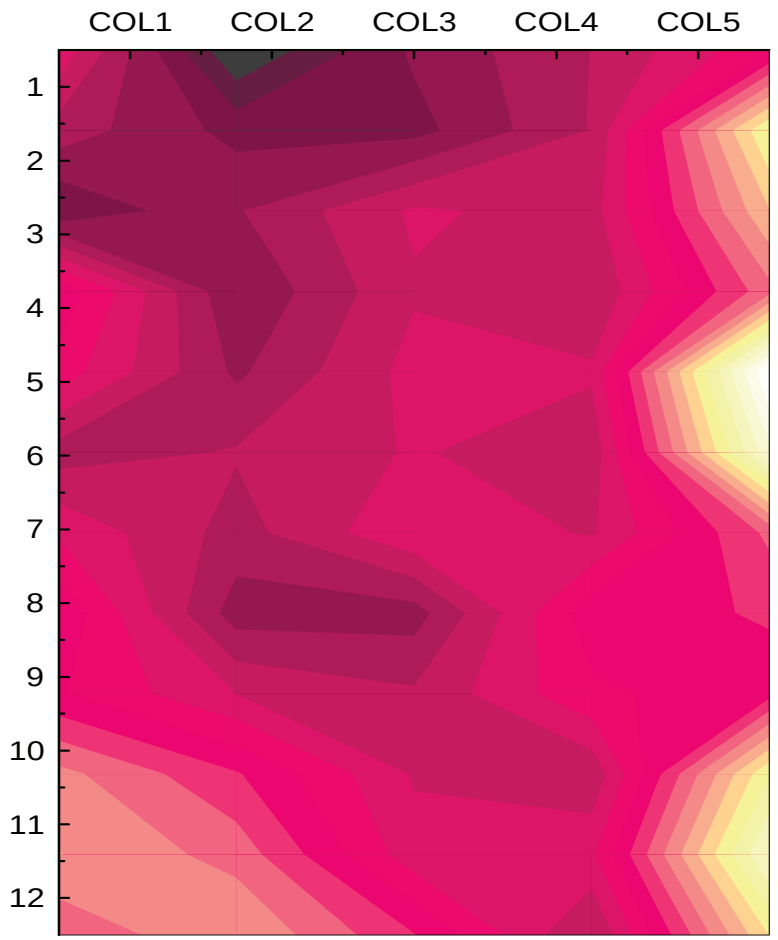


Table A4.4.32. Residual values for grain yield (kg.ha^{-1}) of the Roodebloem 2007 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4	Column 5
			Rep. 1		
1	1493.97	722.27	-670.96	-170.10	-27.58
2	-344.02	407.27	313.40	-81.92	-918.07
3	1509.89	-855.10	-377.91	-201.06	-512.31
4	965.07	700.34	-182.23	343.95	-399.49
			Rep. 2		
5	194.44	-15.31	-631.32	185.45	160.77
6	291.90	338.54	128.02	157.15	-350.02
7	334.14	-320.45	536.40	-242.29	-223.02
8	471.95	426.49	447.83	6.57	-1408.79
			Rep. 3		
9	159.75	-89.71	142.32	-83.86	-456.63
10	250.09	-858.89	10.15	-530.76	-2094.55
11	-814.74	-914.37	137.57	-317.66	-473.57
12	539.94	422.72	1142.14	521.16	531.98

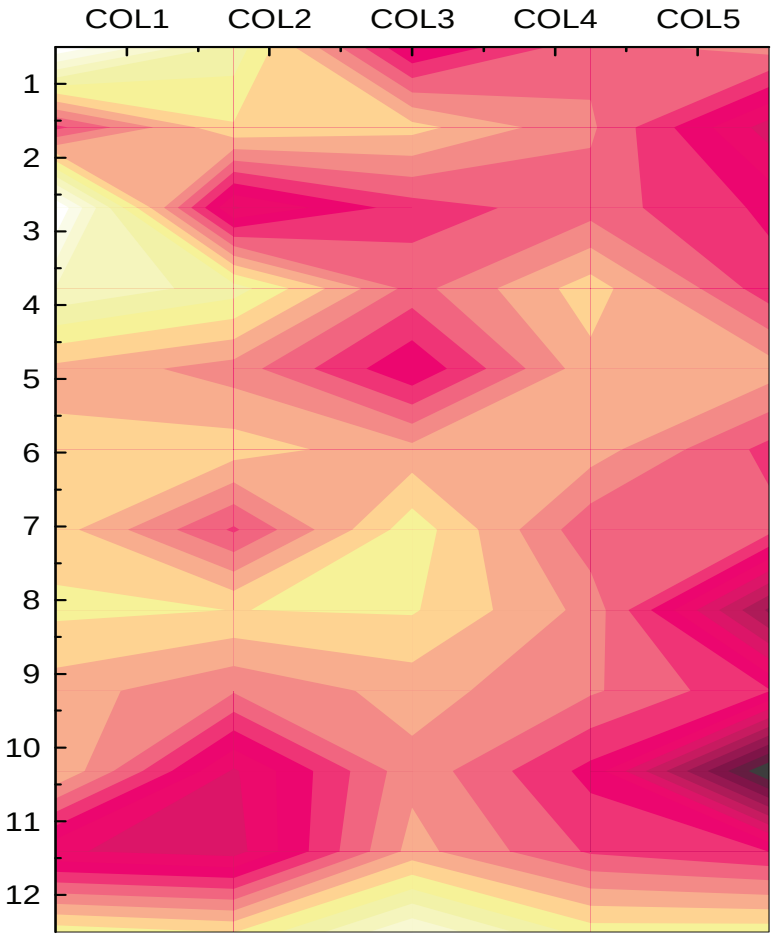


Table A4.4.33. Residual values for grain yield (kg.ha^{-1}) of the Tygerhoek 2007 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4	Column 5
			Rep. 1		
1	58.77	359.01	605.49	346.47	810.04
2	-178.51	-915.63	-80.46	-332.75	184.10
3	-1456.01	-1021.60	443.16	241.45	243.18
4	-440.82	-448.57	288.18	1309.90	52.62
			Rep. 2		
5	555.24	-178.09	748.42	1395.79	493.61
6	-271.82	-339.53	743.04	262.50	237.01
7	274.73	150.89	937.25	876.57	11.90
8	-51.06	-236.98	1059.92	250.73	233.30
			Rep. 3		
9	-218.01	-684.48	174.29	-252.87	427.18
10	-610.90	-521.18	202.02	-580.31	558.87
11	-511.19	-1520.40	238.98	254.86	532.81
12	-579.71	-682.38	-854.23	-258.45	-1040.56

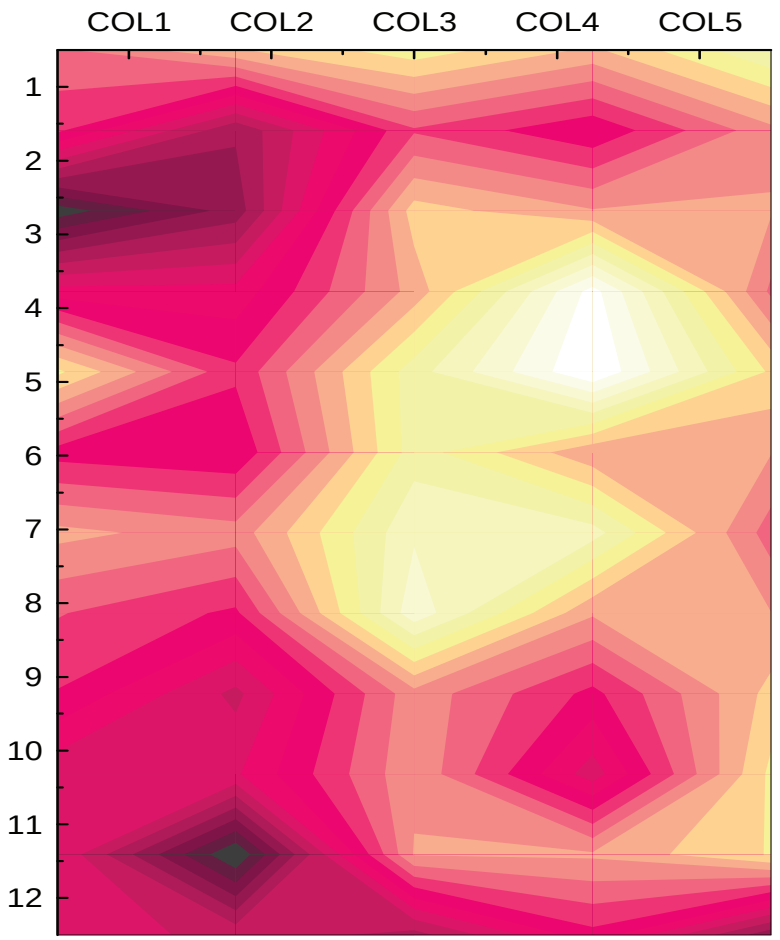


Table A4.4.34. Residual values for grain yield (kg.ha^{-1}) of the Napier 2007 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4
	Rep. 1			
1	570.16	-200.23	-539.68	677.95
2	526.74	-790.35	-355.86	-260.72
3	473.81	-132.72	23.70	705.79
4	600.33	-580.58	-182.00	108.02
5	450.86	-976.84	-873.37	799.48
	Rep. 2			
6	28.90	1237.96	-389.85	620.15
7	-174.64	-562.46	317.70	-1060.56
8	501.98	171.80	-364.77	144.62
9	87.69	520.26	68.29	842.76
10	-220.91	72.80	-0.47	-277.76
	Rep. 3			
11	-286.36	-402.30	-756.37	631.18
12	-313.42	-376.36	-602.23	948.17
13	94.36	-300.19	175.23	709.32
14	-220.28	-297.35	189.44	397.46
15	-57.94	-221.69	-478.02	467.04

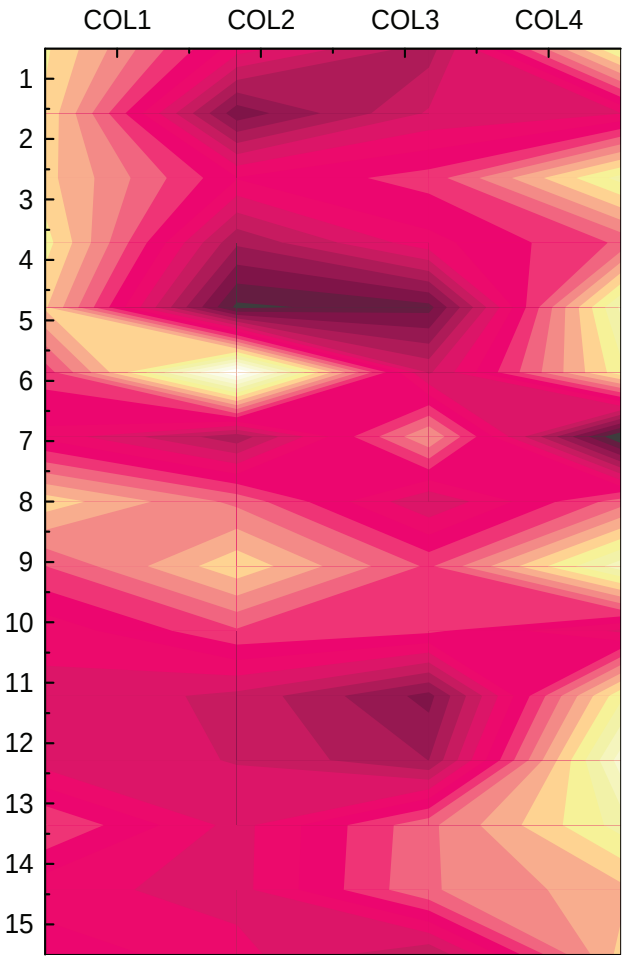


Table A4.4.35. Residual values for grain yield (kg.ha^{-1}) of the Riversdale 2007 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4
	Rep. 3*			
1	1280.80	181.26	19.54	304.79
	Rep. 1			
2	190.94	-1406.34	-1895.38	372.76
3	784.80	-416.76	-420.22	-1643.45
4	1408.94	-677.95	31.18	-1817.96
5	1284.56	1341.05	16.34	-600.11
6	277.52	5.37	-705.87	-883.10
	Rep. 2			
7	2001.10	1112.48	230.14	-570.71
8	980.86	714.38	-733.22	-1591.11
9	1521.68	-30.18	371.20	-631.63
10	264.81	106.76	-453.03	-1687.86
11	-458.28	386.92	1271.07	61.42
	Rep. 3			
12	38.69	-30.95	-145.39	-565.64
13	59.46	793.53	-920.44	-275.18
14	-671.46	31.18	-363.40	-1087.08
15	-791.48	272.14	873.06	1405.53

* Four plots from the 3rd repetition were by mistake planted before the 1st repetition.

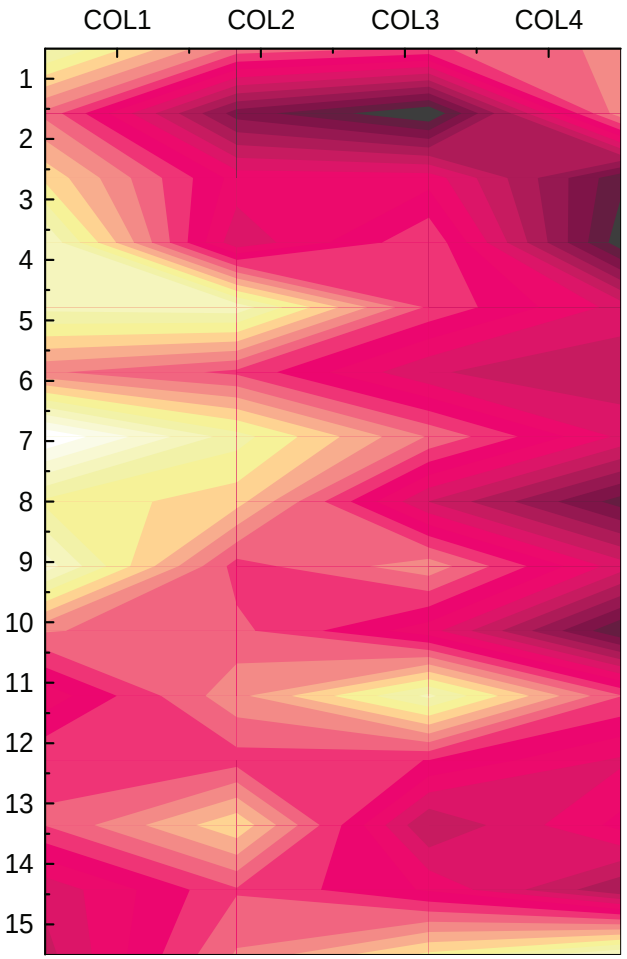
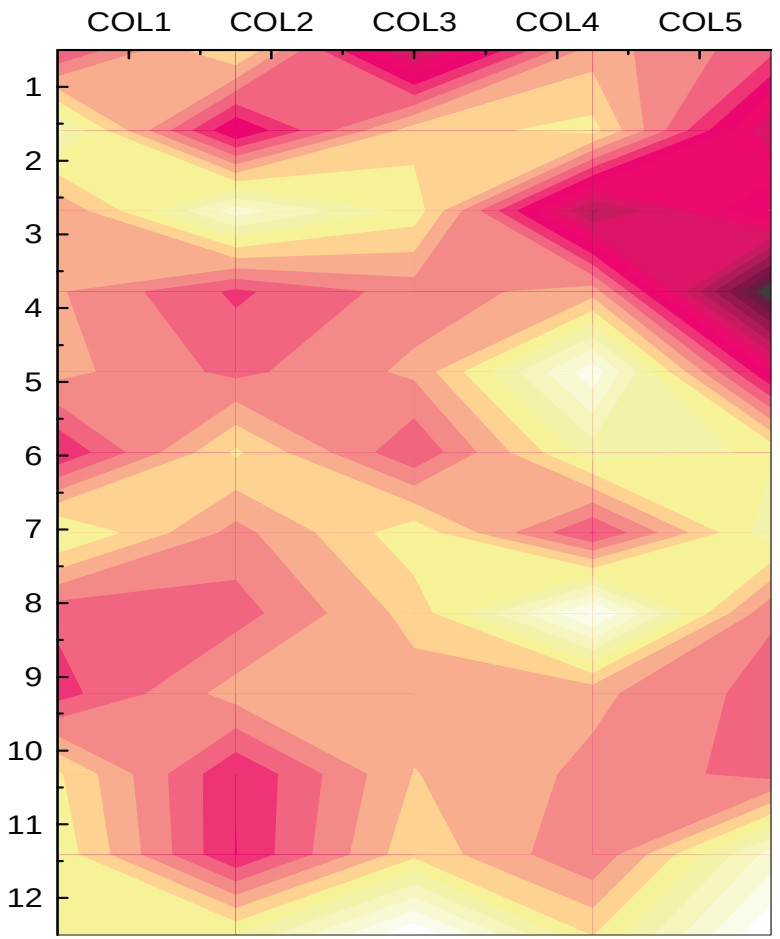


Table A4.4.36. Residual values for grain yield (kg.ha^{-1}) of the Albertinia 2007 elite trial two-dimensional spatial analysis

Row	Column 1	Column 2	Column 3	Column 4	Column 5
			Rep. 1		
1	-335.17	260.81	-949.77	23.13	-277.48
2	549.27	-606.95	167.76	279.24	-852.86
3	-35.33	717.37	314.19	-1038.59	-449.08
4	-28.67	-349.29	-121.59	28.26	-1705.25
			Rep. 2		
5	2.69	-223.49	-14.52	877.67	-624.32
6	-458.63	255.62	-309.09	437.91	350.60
7	409.63	-95.83	325.36	-335.03	459.48
8	-290.47	-247.22	146.93	950.55	-164.17
			Rep. 3		
9	-380.65	18.56	20.19	-12.00	-242.20
10	252.97	-452.75	98.73	-81.50	-243.84
11	327.27	-466.01	192.01	-172.49	701.63
12	347.57	369.31	1042.93	223.05	1060.10



Addendum 5: Cross-site analysis of total starch content, the 2007 season trials

```
PBGXE - CROSS SITE ANALYSIS  FILE 07_ELITE   29/ 8/ 9 20:44
 ------------------------------------------------------------------ :PAGE   1

  20       CN$    CODES:
  1 CA          CA            2 CC          CC            3 CE          CE
  4 CD          CD            5 D1          D1            6 D2          D2
  7 D3          D3            8 D4          D4            9 YC          YC
 10 Y2          Y2           11 EA          EA           12 EB          EB
 13 G1          G1           14 G2          G2           15 H1          H1
 16 H2          H2           17 H3          H3           18 H4          H4
 19 H5          H5           20 H6          H6

   9     SITE$    CODES:
  1 PI 2007       2 KL 2007       3 LA 2007       4 ME 2007       5 RO 2007
  6 TY 2007       7 NA 2007       8 RI 2007       9 AL 2007

 TREATMENT BY VARIATE MEANS HAVE BEEN REQUESTED FOR  1 VARIATES:
 G_STARCH
 ROWS OF MEANS TABLES TO BE SORTED ON VARIATE G_STARCH
 GXE ANALYSIS HAS BEEN REQUESTED FOR  1 VARIATES
```

TREATMENT MEANS AND COUNTS OVER SITES FOR EACH VARIATE. FILE 07_ELITE 29/ 8/ 9 20:44
:PAGE 2

VARIETY\SITE		G_STARCH	
H6	H6	65.54	27
G1	G1	65.33	27
H3	H3	65.27	27
D3	D3	64.63	27
G2	G2	64.47	27
H2	H2	64.24	27
D4	D4	64.18	27
D2	D2	64.12	27
H4	H4	63.98	27
EA	EA	63.92	27
H1	H1	63.82	27
D1	D1	63.59	27
CA	CA	63.54	27
CD	CD	63.42	27
Y2	Y2	63.36	27
CE	CE	63.34	27
H5	H5	63.31	27
CC	CC	62.77	27
EB	EB	62.58	27
YC	YC	62.50	27
SITE MEANS		63.90	540

20 X 9 MATRIX OF TREATMENT BY SITE MEANS FOR VARIATE G_STARCH FILE 07_ELITE 29/ 8/ 9 20:44
:PAGE 3

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
H6	H6	66.65	65.52	66.56	67.53	65.03	62.11	66.67
G1	G1	67.87	65.73	66.35	66.81	67.23	63.04	66.00
H3	H3	67.74	64.17	67.13	66.16	66.00	64.73	63.93
D3	D3	65.42	64.64	64.90	64.93	66.57	62.21	67.46
G2	G2	66.89	63.79	66.04	64.90	64.62	62.52	65.21
H2	H2	65.21	63.82	66.07	65.05	65.10	63.39	65.41
D4	D4	65.11	65.52	65.08	64.15	67.27	62.51	67.43
D2	D2	65.00	64.67	64.35	65.12	67.10	62.45	64.58
H4	H4	64.68	65.52	65.11	64.36	63.96	62.13	66.46
EA	EA	66.15	65.17	65.95	66.62	65.96	61.03	65.12
H1	H1	66.63	63.69	66.19	65.94	65.34	62.65	62.01
D1	D1	67.39	63.00	64.14	63.71	64.93	62.77	63.81
CA	CA	67.56	61.31	66.17	65.83	64.05	62.85	62.69
CD	CD	66.33	63.76	64.36	62.66	66.44	63.08	62.62
Y2	Y2	65.01	61.86	66.30	64.96	65.13	61.66	63.37
CE	CE	66.13	63.54	63.95	64.05	67.14	61.64	63.78
H5	H5	67.05	65.27	66.05	63.85	65.13	59.68	62.00
CC	CC	64.99	65.67	65.13	63.38	62.24	59.92	63.43
EB	EB	61.98	64.89	64.84	63.86	65.12	58.79	63.28
YC	YC	66.35	62.09	64.27	61.35	64.27	60.56	64.68
SITE MEANS		66.01	64.18	65.45	64.76	65.43	61.98	64.50
SITE INDEX		66.01	64.18	65.45	64.76	65.43	61.98	64.50

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	TRT MEANS
H6	H6	65.76	64.06	65.54
G1	G1	61.63	63.30	65.33
H3	H3	64.36	63.23	65.27
D3	D3	63.45	62.14	64.63
G2	G2	62.38	63.88	64.47
H2	H2	61.41	62.66	64.24
D4	D4	59.26	61.28	64.18
D2	D2	62.21	61.62	64.12
H4	H4	61.45	62.11	63.98
EA	EA	59.56	59.76	63.92
H1	H1	59.72	62.19	63.82
D1	D1	60.54	62.03	63.59
CA	CA	59.59	61.82	63.54
CD	CD	59.00	62.53	63.42
Y2	Y2	61.72	60.23	63.36
CE	CE	61.09	58.73	63.34
H5	H5	59.06	61.68	63.31
CC	CC	59.81	60.34	62.77
EB	EB	60.22	60.27	62.58
YC	YC	58.51	60.41	62.50
SITE MEANS		61.04	61.71	63.90
SITE INDEX		61.04	61.71	63.90

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

ENVIRONMENT	MEAN	SE	DUNCAN GROUPS	LSD TESTS
PI 2007	66.007	0.27706	|	.
LA 2007	65.447	0.27706	||	..
RO 2007	65.432	0.27706	||	...
ME 2007	64.761	0.27706	||	2...
NA 2007	64.497	0.27706	|	311..
KL 2007	64.182	0.27706	|	322...
TY 2007	61.985	0.27706	|	333333.
AL 2007	61.713	0.27706	||	333333..
RI 2007	61.037	0.27706	|	3333331..

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

TREATMENT		MEAN	SE	DUNCAN GROUPS	LSD TESTS
H6	H6	65.543	0.41302	|	.
G1	G1	65.329	0.41302	||	..
H3	H3	65.272	0.41302	|||	...
D3	D3	64.635	0.41302	||||	....
G2	G2	64.470	0.41302	||||	.....
H2	H2	64.235	0.41302	||||	1.....
D4	D4	64.179	0.41302	|||	1.....
D2	D2	64.123	0.41302	||||	11.....
H4	H4	63.976	0.41302	|||	211.....
EA	EA	63.925	0.41302	|||	211.....
H1	H1	63.818	0.41302	||||	211.....
D1	D1	63.591	0.41302	||||	222.....
CA	CA	63.540	0.41302	||||	322.....
CD	CD	63.420	0.41302	||||	3221.....
Y2	Y2	63.360	0.41302	||||	3321.....
CE	CE	63.338	0.41302	||||	3321.....
H5	H5	63.308	0.41302	||||	33311.....
CC	CC	62.769	0.41302	|||	3332211111.....
EB	EB	62.583	0.41302	||	33332222111.....
YC	YC	62.498	0.41302	|	33333222111.....

RESIDUALS FROM THE ADDITIVE TREATMENT BY SITE MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN STANDARD ERRORS,
 ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		P I	L A	R O	M E	N A	K L	T Y	A L	R I	T - E
		2	2	2	2	2	2	2	2	2	F
		0	0	0	0	0	0	0	0	0	C
		0	0	0	0	0	0	0	0	0	T
		7	7	7	7	7	7	7	7	7	S
H6	H6	0	0	-1	0	0	0	-1	0	2	6
G1	G1	0	0	0	0	0	0	0	0	0	3
H3	H3	0	0	0	0	-1	-1	1	0	1	3
D3	D3	-1	-1	0	0	1	0	0	0	1	1
G2	G2	0	0	-1	0	0	0	0	1	0	1
H2	H2	0	0	0	0	0	0	0	0	0	0
D4	D4	-1	0	1	0	2	0	0	0	-1	0
D2	D2	-1	-1	1	0	0	0	0	0	0	0
H4	H4	-1	0	-1	0	1	1	0	0	0	0
EA	EA	0	0	0	1	0	0	0	-1	-1	0
H1	H1	0	0	0	1	-2	0	0	0	-1	0
D1	D1	1	0	0	0	0	0	0	0	0	0
CA	CA	1	0	0	1	-1	-2	1	0	0	0
CD	CD	0	0	1	-1	-1	0	1	1	-1	-1
Y2	Y2	0	1	0	0	0	-1	0	0	1	-1
CE	CE	0	0	1	0	0	0	0	-2	0	-1
H5	H5	1	1	0	0	-1	1	-1	0	-1	-1
CC	CC	0	0	-1	0	0	2	0	0	0	-2
EB	EB	-2	0	0	0	0	1	-1	0	0	-3
YC	YC	1	0	0	-1	1	0	0	0	0	-3
SITE EFFECTS		8	5	5	3	2	1	-7	0	-10	691

```
BOX PLOT OF  180 STUDENTIZED RESIDUALS FROM LPLT= -2.382    TO ULPT=  2.702
NO.<LPLT                                                          NO.>UPLT
       0 -------------------I        +      I--------------------   *   0

MEDIAN=  0.5735E-01 ANDERSON-DARLING STATISTIC=  0.262

ANALYSIS OF VARIANCE FOR THE ADDITIVE MODEL

-------------------------------------------------------------------
SOURCE                  D.F.     S.S.         M.S.     F       FPROB
-------------------------------------------------------------------
TREATMENTS               19    127.171        6.69319
LOCATIONS                 8    540.058        67.5073
TREATMENT X SITES       152    233.358        1.53525
-------------------------------------------------------------------
TOTAL                   179    900.587
```

RESIDUALS FROM THE ADDITIVE SITE X TREATMENT MODEL FILE 07_ELITE   29/ 8/ 9 20:44
 ---------- :PAGE   5

 SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
H6	H6	-1.007	-.3046	-.5336	1.120	-2.051	-1.526	0.5214
G1	G1	0.4292	0.1146	-.5292	0.6199	0.3690	-.3826	0.6803E-01
H3	H3	0.3534	-1.385	0.3032	0.2228E-01	-.8085	1.371	-1.941
D3	D3	-1.331	-.2794	-1.290	-.5680	0.3955	-.5110	2.222
G2	G2	0.3058	-.9684	0.1792E-01	-.4396	-1.385	-.3867E-01	0.1408
H2	H2	-1.139	-.6984	0.2866	-.5503E-01	-.6718	1.069	0.5711
D4	D4	-1.181	1.059	-.6546	-.8887	1.552	0.2385	2.652 *
D2	D2	-1.234	0.2611	-1.324	0.1285	1.444	0.2418	-.1428
H4	H4	-1.405	1.258	-.4160	-.4805	-1.549	0.6258E-01	1.885
EA	EA	0.1104	0.9635	0.4772	1.828	0.5016	-.9855	0.5934
H1	H1	0.7048	-.4094	0.8165	1.262	-.1823E-01	0.7381	-2.410 *
D1	D1	1.688	-.8797	-1.005	-.7479	-.1937	1.088	-.3774
CA	CA	1.912	-2.521 *	1.077	1.421	-1.023	1.218	-1.453
CD	CD	0.8032	0.5589E-01	-.6119	-1.624	1.480	1.573	-1.402
Y2	Y2	-.4581	-1.790	1.392	0.7317	0.2298	0.2077	-.5918
CE	CE	0.6757	-.8957E-01	-.9360	-.1502	2.269 *	0.2093	-.1584
H5	H5	1.628	1.679	1.194	-.3234	0.2811	-1.720	-1.906
CC	CC	0.1095	2.619 *	0.8122	-.2556	-2.062	-.9386	0.5962E-01
EB	EB	-2.712 *	2.016	0.7052	0.4123	1.001	-1.884	0.9210E-01
YC	YC	1.745	-.6985	0.2177	-2.013	0.2394	-.3081E-01	1.577
SITE EFFECTS		2.111 ***	0.2866	1.551 ***	0.8650 **	1.536 ***	-1.911 ***	0.6015 *

SECTION 2

VARIETY\SITE		RI 2007		AL 2007		T-EFCTS	
H6	H6	3.076	**	0.7047		1.647	***
G1	G1	-.8418		0.1528		1.433	***
H3	H3	1.942		0.1435		1.376	***
D3	D3	1.677		-.3154		0.7393	
G2	G2	0.7728		1.594		0.5748	
H2	H2	0.3044E-01		0.6065		0.3394	
D4	D4	-2.056		-.7205		0.2830	
D2	D2	0.9433		-.3170		0.2272	
H4	H4	0.3304		0.3132		0.8064E-01	
EA	EA	-1.505		-1.984		0.2916E-01	
H1	H1	-1.238		0.5549		-.7803E-01	
D1	D1	-.1916		0.6181		-.3047	
CA	CA	-1.090		0.4585		-.3557	
CD	CD	-1.563		1.289		-.4755	
Y2	Y2	1.222		-.9437		-.5360	
CE	CE	0.6083		-2.428	*	-.5573	
H5	H5	-1.389		0.5551		-.5876	
CC	CC	-.9899E-01		-.2451		-1.126	**
EB	EB	0.4992		-.1291		-1.312	**
YC	YC	-1.129		0.9244E-01		-1.397	***
SITE EFFECTS		-2.858	***	-2.183		63.90	***

FITTED VALUES FROM ADDITIVE SITE X TREATMENT MODEL FILE 07_ELITE   29/ 8/ 9 20:44
 -- :PAGE   6

 SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
H6	H6	67.65	65.83	67.09	66.41	67.08	63.63	66.14
G1	G1	67.44	65.62	66.88	66.19	66.87	63.42	65.93
H3	H3	67.38	65.56	66.82	66.14	66.81	63.36	65.87
D3	D3	66.75	64.92	66.19	65.50	66.17	62.72	65.24
G2	G2	66.58	64.76	66.02	65.34	66.01	62.56	65.07
H2	H2	66.35	64.52	65.79	65.10	65.77	62.32	64.84
D4	D4	66.29	64.47	65.73	65.04	65.71	62.27	64.78
D2	D2	66.23	64.41	65.67	64.99	65.66	62.21	64.72
H4	H4	66.09	64.26	65.53	64.84	65.51	62.07	64.58
EA	EA	66.04	64.21	65.48	64.79	65.46	62.01	64.53
H1	H1	65.93	64.10	65.37	64.68	65.35	61.91	64.42
D1	D1	65.70	63.88	65.14	64.46	65.13	61.68	64.19
CA	CA	65.65	63.83	65.09	64.40	65.08	61.63	64.14
CD	CD	65.53	63.71	64.97	64.29	64.96	61.51	64.02
Y2	Y2	65.47	63.65	64.91	64.22	64.90	61.45	63.96
CE	CE	65.45	63.62	64.89	64.20	64.87	61.43	63.94
H5	H5	65.42	63.59	64.86	64.17	64.84	61.40	63.91
CC	CC	64.88	63.06	64.32	63.63	64.31	60.86	63.37
EB	EB	64.69	62.87	64.13	63.45	64.12	60.67	63.18
YC	YC	64.61	62.78	64.05	63.36	64.03	60.59	63.10
SITE ESTS.		66.01	64.18	65.45	64.76	65.43	61.98	64.50

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	T-ESTS.
H6	H6	62.68	63.36	65.54
G1	G1	62.47	63.15	65.33
H3	H3	62.41	63.09	65.27
D3	D3	61.78	62.45	64.63
G2	G2	61.61	62.29	64.47
H2	H2	61.38	62.05	64.24
D4	D4	61.32	62.00	64.18
D2	D2	61.26	61.94	64.12
H4	H4	61.12	61.79	63.98
EA	EA	61.07	61.74	63.92
H1	H1	60.96	61.63	63.82
D1	D1	60.73	61.41	63.59
CA	CA	60.68	61.36	63.54
CD	CD	60.56	61.24	63.42
Y2	Y2	60.50	61.18	63.36
CE	CE	60.48	61.16	63.34
H5	H5	60.45	61.13	63.31
CC	CC	59.91	60.59	62.77
EB	EB	59.73	60.40	62.58
YC	YC	59.64	60.32	62.50
SITE ESTS.		61.04	61.71	63.90

 REGRESSIONS OF G_STARCH FOR EACH VARIETY ON MEANS OF G_STARCH AT EACH SITE

VARIETY		MEAN	SLOPE	SE	MS-TXL	MS-REG	MS-DEV	R**2(%)
CA	CA	63.54	1.132	0.315	2.40	0.47	2.68	2.
CC	CC	62.77	1.056	0.259	1.60	0.08	1.82	1.
CE	CE	63.34	1.236	0.245	1.61	1.50	1.62	12.
CD	CD	63.42	0.979	0.276	1.80	0.01	2.06	0.
D1	D1	63.59	0.915	0.189	0.87	0.20	0.97	3.
D2	D2	64.12	0.822	0.173	0.81	0.86	0.81	13.
D3	D3	64.63	0.757	0.236	1.51	1.60	1.50	13.
D4	D4	64.18	1.259	0.292	2.25	1.81	2.31	10.
YC	YC	62.50	1.240	0.229	1.43	1.56	1.41	14.
Y2	Y2	63.36	0.981	0.215	1.09	0.01	1.24	0.
EA	EA	63.92	1.536*	0.155	1.54	7.76	0.65	63.
EB	EB	62.58	1.013	0.299	2.11	0.00	2.42	0.
G1	G1	65.33	1.151	0.082	0.24	0.61	0.18	33.
G2	G2	64.47	0.717	0.148	0.79	2.16	0.59	34.
H1	H1	63.82	1.117	0.238	1.39	0.37	1.54	3.
H2	H2	64.24	0.765	0.119	0.52	1.49	0.38	36.
H3	H3	65.27	0.628	0.212	1.53	3.74	1.21	31.
H4	H4	63.98	0.753	0.212	1.27	1.64	1.21	16.
H5	H5	63.31	1.401	0.247	1.99	4.33	1.65	27.
H6	H6	65.54	0.544	0.270	2.43	5.62	1.97	29.

 SLOPE - SLOPES OF REGRESSIONS OF VARIETY MEANS ON SITE INDEX.
          * INDICATES SLOPES SIGNIFICANTLY DIFFERENT FROM THE
          SLOPE FOR THE OVERALL REGRESSION WHICH IS   1.00
 MS-TXL - CONTRIBUTION OF EACH VARIETY TO INTERACTION MS
 MS-REG - CONTRIBUTION OF EACH VARIETY TO THE REGRESSION
          COMPONENT OF THE TREATMENT BY LOCATION INTERACTION
 MS-DEV - DEVIATIONS FROM REGRESSION COMPONENT OF INTERACTION
 R**2   - SQUARED CORRELATION BETWEEN RESIDUALS FROM THE MAIN
          EFFECTS MODEL AND THE SITE INDEX.

```
VARIATE G_STARCH WAS SITE INDEX WITH OVERALL MEAN  63.90
THE FOLLOWING SITE MEANS OF G_STARCH WERE USED AS X-VARIATES
          66.01     64.18     65.45     64.76     65.43     61.98     64.50     61.04     61.71
-------------------------------------------------------------------
ANOVA FOR VARIABLE G_STARCH WITH SITE REGRESSIONS ON G_STARCH
-------------------------------------------------------------------
SOURCE             D.F.      S.S.         M.S.     F       FPROB
-------------------------------------------------------------------
TREATMENTS          19    127.171        6.69319
LOCATIONS            8    540.058        67.5073
TREATMENT X SITES  152    233.358        1.53525
   TRT X SITE REG   19    35.8439        1.88652       1.270   0.213
   DEVIATIONS      133    197.514        1.48507
-------------------------------------------------------------------
TOTAL              179    900.587
```

```
AMMI ANALYSIS  FILE 07_ELITE   29/ 8/ 9 20:44
 ------------------------------------------------------------ :PAGE   8
 SINGULAR VALUES OF INTERACTION MATRIX (CONDITION=  0)
 8.4303    7.2099    6.1140    5.4161    4.1912    3.6130    2.5916    2.5008

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR TREATMENTS

 CA CA          CA          -0.135798E+01 0.240071E+00-0.637826E-01 0.512823E-01
 CC CC          CC           0.393930E+00 0.133133E-01-0.103209E+01-0.707183E+00
 CE CE          CE           0.112706E+00-0.492289E+00 0.521554E+00 0.108735E+01
 CD CD          CD          -0.672910E+00-0.779835E+00 0.468509E+00-0.279048E+00
 D1 D1          D1          -0.620053E+00-0.603238E-01 0.554811E+00-0.369660E+00
 D2 D2          D2           0.406828E+00 0.213439E-01 0.395704E+00 0.628515E+00
 D3 D3          D3           0.923878E+00 0.378541E+00 0.749482E+00 0.421973E-01
 D4 D4          D4           0.857173E+00-0.106718E+01 0.547087E+00-0.203737E+00
 YC YC          YC          -0.169145E+00-0.584024E+00 0.595346E+00-0.762806E+00
 Y2 Y2          Y2          -0.260009E+00 0.553195E+00 0.718201E-01 0.789105E+00
 EA EA          EA           0.314205E+00-0.621826E+00-0.641012E+00 0.594294E+00
 EB EB          EB           0.104327E+01-0.526199E-01-0.766787E+00 0.459694E+00
 G1 G1          G1          -0.469914E-01-0.289061E+00-0.901305E-01 0.319041E-01
 G2 G2          G2          -0.238938E+00 0.632226E+00 0.144106E+00-0.661424E+00
 H1 H1          H1          -0.931232E+00-0.132219E+00-0.551617E+00 0.296120E+00
 H2 H2          H2           0.338326E-01 0.330369E+00 0.306271E+00-0.241185E+00
 H3 H3          H3          -0.709185E+00 0.867735E+00 0.190780E+00 0.319879E+00
 H4 H4          H4           0.801812E+00 0.272687E+00 0.278789E-01-0.746701E+00
 H5 H5          H5          -0.421924E+00-0.674572E+00-0.115312E+01-0.237989E+00
 H6 H6          H6           0.540726E+00 0.144447E+01-0.274809E+00-0.906120E-01

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR ENVIRONMENTS

 PI             PI 2007     -0.156300E+01-0.568322E+00 0.128724E-02-0.442640E+00
 KL             KL 2007      0.123367E+01-0.872070E+00-0.122730E+01-0.420215E+00
 LA             LA 2007     -0.485031E+00 0.670447E-01-0.101436E+01 0.198676E-01
 ME             ME 2007     -0.157796E+00 0.549893E+00-0.875357E+00 0.101278E+01
 RO             RO 2007      0.208013E+00-0.136813E+01 0.713628E+00 0.129201E+01
 TY             TY 2007     -0.933843E+00-0.292054E-01 0.112403E+01-0.289739E-01
 NA             NA 2007      0.163631E+01-0.183488E+00 0.955556E+00-0.763287E+00
 RI             RI 2007      0.583342E+00 0.191731E+01 0.354627E+00 0.542884E+00
 AL             AL 2007     -0.521668E+00 0.486968E+00-0.321102E-01-0.121243E+01
```

RESIDUALS FROM THE AMMI-2 MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN UNITS OF ROOT (RESIDUAL GXE MS), ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		P I 2 0 0 7	L A 2 0 0 7	R O 2 0 0 7	M E 2 0 0 7	N A 2 0 0 7	K L 2 0 0 7	T Y 2 0 0 7	A L 2 0 0 7	R I 2 0 0 7	T - E F C T S
H6	H6	0	0	0	0	0	0	0	0	0	EA
G1	G1	0	0	0	0	0	0	0	0	0	EA
H3	H3	0	0	0	0	0	0	0	0	0	EA
D3	D3	0	0	0	0	0	-1	0	0	0	EA
G2	G2	0	0	0	0	0	0	0	1	0	EA
H2	H2	0	0	0	0	0	0	1	0	0	EA
D4	D4	0	0	0	0	1	0	0	0	0	EA
D2	D2	0	-1	1	0	0	0	0	0	0	EA
H4	H4	0	0	-1	0	0	0	0	0	0	EA
EA	EA	0	0	0	2	0	0	0	-1	0	EA
H1	H1	0	0	0	1	0	0	0	0	0	EA
D1	D1	0	-1	0	0	0	0	0	0	0	EA
CA	CA	0	0	0	1	0	0	0	0	0	EA
CD	CD	0	0	0	-1	0	0	0	1	0	EA
Y2	Y2	0	1	1	0	0	0	0	-1	0	EA
CE	CE	0	0	1	0	0	0	0	-2	1	EA
H5	H5	0	0	0	0	-1	1	-2	0	0	EA
CC	CC	0	0	-2	0	0	2	0	0	0	EA
EB	EB	-1	1	0	0	-1	0	0	0	0	EA
YC	YC	1	0	0	-1	1	0	0	0	0	EA

```
BOX PLOT OF  180 STANDERSIZED RESIDUALS FROM LPLT= -2.051    TO ULPT=  2.134
NO.<LPLT                                                          NO.>UPLT
       0 ----------------------I      +       I----------------------   0
```

ANALYSIS OF VARIANCE FOR THE AMMI MODEL

SOURCE	D.F.	S.S.	M.S.	F	FPROB
TREATMENTS	19	127.171	6.69319		
LOCATIONS	8	540.058	67.5073		
TREATMENT X SITES	152	233.358	1.53525		
AMMI COMPONENT 1	26	71.0701	2.73347	2.122	0.003
AMMI COMPONENT 2	24	51.9820	2.16592	2.003	0.009
AMMI COMPONENT 3	22	37.3814	1.69916	1.864	0.024
AMMI COMPONENT 4	20	29.3338	1.46669	2.019	0.019
GXE RESIDUAL	60	43.5904			
TOTAL	179	900.587			

 GENOTYPE MAP SHOWING BEST GENOTYPES OVER THE RANGE OF AMMI-2 SITE SCORES (2 CHRS/PIXEL)

```
 1.92 H3H3H3H3H3H3H3H3H6H6H6
```

```
-0.31 H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6D3D3D3
-0.38 H3H3H3H3H3H3H3H3H3H3H3H3H3H3G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1H6H6H6H6H6H6H6H6H6D3D3D3D3D3D3
-0.44 H3H3H3H3H3H3H3H3H3H3H3H3G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4
-0.50 H3H3H3H3H3H3H3H3H3H3G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-0.56 H3H3H3H3H3H3H3H3H3G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-0.62 H3H3H3H3H3H3H3G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-0.69 H3H3H3H3H3G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-0.75 H3H3H3G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-0.81 H3H3G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-0.87 G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-0.93 G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-1.00 G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-1.06 G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-1.12 G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-1.18 G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-1.24 G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-1.31 G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
-1.37 G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1G1D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4D4
       ^           ^           ^           ^           ^           ^           ^           ^           ^
   -0.156E+01  -0.120E+01  -0.839E+00  -0.476E+00  -0.114E+00   0.248E+00   0.610E+00   0.972E+00   0.133E+01
```

AMMI RESIDUALS, ADDITIVE EFFECTS AND MULTIPLICATIVE SCORES FILE 07_ELITE   29/ 8/ 9 20:44
:PAGE 10

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
H6	H6	0.6593	0.2880	-.3681	0.4115	-.1876	-.9793	-.9830E-01
G1	G1	0.1915	-.7954E-01	-.5326	0.7715	-.1674E-01	-.4349	0.9188E-01
H3	H3	-.2619	0.2462	-.9894E-01	-.5668	0.5262	0.7336	-.6217
D3	D3	0.3282	-1.089	-.8670	-.6303	0.7212	0.3628	0.7795
G2	G2	0.2917	-.1222	-.1404	-.8249	-.4703	-.2433	0.6478
H2	H2	-.8979	-.4520	0.2808	-.2314	-.2269	1.110	0.5763
D4	D4	-.4481	-.9295	-.1673	-.1666	-.8628E-01	1.008	1.054
D2	D2	-.5862	-.2222	-1.129	0.1810	1.388	0.6224	-.8046
H4	H4	0.3452E-02	0.5069	-.4541E-01	-.5040	-1.342	0.8193	0.6235
EA	EA	0.2481	0.3357E-01	0.6713	2.219 *	-.4145	-.7103	-.3480E-01
H1	H1	-.8258	0.6241	0.3737	1.187	-.5419E-02	-.1353	-.9107
D1	D1	0.6849	-.1673	-1.301	-.8125	-.1472	0.5076	0.6261
CA	CA	-.7362E-01	-.6368	0.4021	1.075	-.4124	-.4335E-01	0.8136
CD	CD	-.6917	0.2060	-.8860	-1.302	0.5529	0.9222	-.4438
Y2	Y2	-.5501	-.9866	1.229	0.3864	1.041	-.1894E-01	-.6484E-01
CE	CE	0.5721	-.6579	-.8483	0.1383	1.572	0.3002	-.4332
H5	H5	0.5856	1.611	1.035	-.1902E-01	-.5540	-2.133 *	-1.339
CC	CC	0.7328	2.144 *	1.002	-.2008	-2.125 *	-.5703	-.5825
EB	EB	-1.111	0.6827	1.215	0.6059	0.7117	-.9116	-1.625
YC	YC	1.149	-.9991	0.1748	-1.718	-.5244	-.2058	1.746
SITE EFFECTS		2.111	0.2866	1.551	0.8650 ***	1.536 ***	-1.911 ***	0.6015
AMMI1 SITE		-1.563	1.234 ***	-.4850 ***	-.1578 ***	0.2080 ***	-.9338 ***	1.636 ***
AMMI2 SITE		-.5683	-.8721	0.6704E-01	0.5499	-1.368 ***	-.2921E-01	-.1835

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	T-EFCTS	AMMI1 TRT	AMMI2 TRT
H6	H6	-.8888E-02	0.2834	1.647 ***	0.5407	1.444 ***
G1	G1	-.2602	0.2691	1.433 ***	-.4699E-01	-.2891 ***
H3	H3	0.6924	-.6490	1.376	-.7092	0.8677
D3	D3	0.4124	-.1782E-01	0.7393 ***	0.9239 ***	0.3785
G2	G2	-.3000	1.162	0.5748 ***	-.2389 ***	0.6322 ***
H2	H2	-.6227	0.4633	0.3394	0.3383E-01	0.3304 ***
D4	D4	-.5099	0.2463	0.2830	0.8572 ***	-1.067
D2	D2	0.6651	-.1152	0.2272 ***	0.4068 ***	0.2134E-01
H4	H4	-.6601	0.5987	0.8064E-01	0.8018 ***	0.2727 ***
EA	EA	-.4958	-1.517	0.2916E-01	0.3142 ***	-.6218
H1	H1	-.4416	0.1335	-.7803E-01	-.9312	-.1322
D1	D1	0.2857	0.3240	-.3047	-.6201	-.6032E-01
CA	CA	-.7578	-.3668	-.3557	-1.358 ***	0.2401
CD	CD	0.3245	1.318	-.4755	-.6729	-.7798 ***
Y2	Y2	0.3131	-1.349	-.5360 ***	-.2600 ***	0.5532
CE	CE	1.486	-2.130 *	-.5573 ***	0.1127 ***	-.4923 ***
H5	H5	0.1509	0.6635	-.5876 ***	-.4219	-.6746
CC	CC	-.3543	-.4608E-01	-1.126	0.3939 ***	0.1331E-01
EB	EB	-.8504E-02	0.4407	-1.312	1.043	-.5262E-01
YC	YC	0.8928E-01	0.2886	-1.397	-.1691	-.5840 ***
SITE EFFECTS		-2.858	-2.183 ***	63.90 ***	.	.
AMMI1 SITE		0.5833 ***	-.5217 ***	.	.	.
AMMI2 SITE		1.917 ***	0.4870	.	.	.

FITTED VALUES FROM THE AMMI MODEL FILE 07_ELITE   29/ 8/ 9 20:44
---------- :PAGE 11

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
H6	H6	65.99	65.24	66.93	67.12	65.22	63.08	66.76
G1	G1	67.68	65.81	66.88	66.04	67.25	63.47	65.91
H3	H3	68.00	63.93	67.23	66.73	65.47	64.00	64.55
D3	D3	65.09	65.73	65.76	65.56	65.85	61.85	66.68
G2	G2	66.60	63.91	66.18	65.72	65.09	62.76	64.56
H2	H2	66.11	64.28	65.79	65.28	65.33	62.28	64.83
D4	D4	65.56	66.45	65.24	64.32	67.35	61.50	66.38
D2	D2	65.59	64.89	65.48	64.94	65.71	61.83	65.39
H4	H4	64.68	65.01	65.16	64.86	65.31	61.31	65.84
EA	EA	65.90	65.14	65.28	64.40	66.38	61.74	65.15
H1	H1	67.46	63.07	65.81	64.76	65.34	62.78	62.92
D1	D1	66.71	63.17	65.44	64.52	65.08	62.26	63.19
CA	CA	67.64	61.94	65.77	64.75	64.47	62.89	61.88
CD	CD	67.03	63.56	65.25	63.96	65.88	62.16	63.06
Y2	Y2	65.56	62.84	65.07	64.57	64.08	61.68	63.43
CE	CE	65.55	64.19	64.80	63.91	65.57	61.34	64.21
H5	H5	66.46	63.66	65.02	63.87	65.68	61.81	63.34
CC	CC	64.26	63.53	64.13	63.58	64.37	60.49	64.01
EB	EB	63.09	64.20	63.63	63.25	64.41	59.70	64.90
YC	YC	65.21	63.09	64.09	63.07	64.80	60.76	62.93
SITE ESTS.		66.01	64.18	65.45	64.76	65.43	61.98	64.50
AMMI1 SITE		-1.563	1.234	-.4850	-.1578	0.2080	-.9338	1.636
AMMI2 SITE		-.5683	-.8721	0.6704E-01	0.5499	-1.368	-.2921E-01	-.1835

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	T-ESTS.	AMMI1 TRT	AMMI2 TRT
H6	H6	65.77	63.78	65.54	0.5407	1.444 I
G1	G1	61.89	63.03	65.33	-.4699E-01	-.2891 I
H3	H3	63.66	63.88	65.27	-.7092	0.8677 I
D3	D3	63.04	62.15	64.63	0.9239	0.3785 I
G2	G2	62.68	62.72	64.47	-.2389	0.6322 I
H2	H2	62.03	62.20	64.24	0.3383E-01	0.3304 I
D4	D4	59.77	61.03	64.18	0.8572	-1.067 I
D2	D2	61.54	61.74	64.12	0.4068	0.2134E-01 I
H4	H4	62.11	61.51	63.98	0.8018	0.2727 I
EA	EA	60.06	61.28	63.92	0.3142	-.6218 I
H1	H1	60.16	62.06	63.82	-.9312	-.1322 I
D1	D1	60.26	61.70	63.59	-.6201	-.6032E-01 I
CA	CA	60.35	62.18	63.54	-1.358	0.2401 I
CD	CD	58.67	61.21	63.42	-.6729	-.7798 I
Y2	Y2	61.41	61.58	63.36	-.2600	0.5532 I
CE	CE	59.60	60.86	63.34	0.1127	-.4923 I
H5	H5	58.91	61.02	63.31	-.4219	-.6746 I
CC	CC	60.17	60.39	62.77	0.3939	0.1331E-01 I
EB	EB	60.23	59.83	62.58	1.043	-.5262E-01 I
YC	YC	58.42	60.12	62.50	-.1691	-.5840 I
SITE ESTS.		61.04	61.71	63.90	.	.
AMMI1 SITE		0.5833	-.5217	.	.	.
AMMI2 SITE		1.917 I	0.4870 I	.	.	.

Addendum 6: Cross-site analysis of total starch yield, the 2006 season trials

```
PBGXE - CROSS SITE ANALYSIS  FILE 06_ELITE   15/ 7/ 9 22:48
 ------------------------------------------------------------------ :PAGE   1

  20       CN$    CODES:
  1 CA          CA            2 CB          CB            3 CC          CC
  4 CD          CD            5 CE          CE            6 D1          D1
  7 DB          DB            8 YA          YA            9 DC          DC
 10 YB          YB           11 YC          YC           12 DD          DD
 13 DE          DE           14 DF          DF           15 DG          DG
 16 DH          DH           17 Y1          Y1           18 Y2          Y2
 19 YD          YD           20 Y3          Y3

   6     SITE$    CODES:
  1 VR 2006       2 LA 2006       3 ME 2006       4 RO 2006       5 TY 2006
  6 NA 2006

 TREATMENT BY VARIATE MEANS HAVE BEEN REQUESTED FOR  1 VARIATES:
 STY_KGHA
 ROWS OF MEANS TABLES TO BE SORTED ON VARIATE STY_KGHA
 GXE ANALYSIS HAS BEEN REQUESTED FOR  1 VARIATES
```

TREATMENT MEANS AND COUNTS OVER SITES FOR EACH VARIATE. FILE 06_ELITE 15/ 7/ 9 22:48
:PAGE 2

VARIETY\SITE		STY_KGHA	
CA	CA	2395.	24
Y3	Y3	2379.	24
YC	YC	2336.	24
CC	CC	2289.	24
Y1	Y1	2275.	24
YA	YA	2274.	24
YB	YB	2264.	24
Y2	Y2	2260.	24
DC	DC	2237.	24
DB	DB	2201.	24
YD	YD	2200.	24
DE	DE	2184.	24
CE	CE	2174.	24
DH	DH	2149.	24
DF	DF	2137.	24
DD	DD	2126.	24
D1	D1	2124.	24
CD	CD	2089.	24
DG	DG	1945.	24
CB	CB	1889.	24
SITE MEANS		2196.	480

20 X 6 MATRIX OF TREATMENT BY SITE MEANS FOR VARIATE STY_KGHA FILE 06_ELITE 15/ 7/ 9 22:48

VARIETY\SITE		VR 2006	LA 2006	ME 2006	RO 2006	TY 2006	NA 2006	TRT MEANS
CA	CA	1786.	2017.	1638.	3577.	2251.	3104.	2395.
Y3	Y3	1506.	1863.	1876.	3116.	2523.	3391.	2379.
YC	YC	2098.	1536.	1407.	3489.	2633.	2852.	2336.
CC	CC	1880.	1599.	1313.	3228.	2301.	3412.	2289.
Y1	Y1	1411.	1917.	1517.	3351.	2449.	3004.	2275.
YA	YA	1732.	1924.	1511.	3189.	2358.	2930.	2274.
YB	YB	2448.	1886.	1481.	3334.	2546.	1888.	2264.
Y2	Y2	1584.	1735.	1494.	3001.	2667.	3081.	2260.
DC	DC	1810.	2187.	1620.	2815.	2243.	2745.	2237.
DB	DB	1787.	1491.	1584.	3366.	2102.	2873.	2201.
YD	YD	1515.	1973.	1385.	2895.	2538.	2894.	2200.
DE	DE	1670.	1745.	1613.	3080.	2288.	2705.	2184.
CE	CE	1665.	1604.	1696.	3258.	2124.	2695.	2174.
DH	DH	1421.	1647.	1447.	3391.	2136.	2853.	2149.
DF	DF	1766.	1566.	1554.	3061.	2338.	2535.	2137.
DD	DD	2011.	1558.	1527.	2942.	2073.	2642.	2126.
D1	D1	1670.	1384.	1706.	3147.	1952.	2886.	2124.
CD	CD	2186.	2007.	1161.	3213.	1566.	2399.	2089.
DG	DG	1766.	1528.	1273.	2875.	2111.	2117.	1945.
CB	CB	1793.	1152.	945.9	2749.	2188.	2508.	1889.
SITE MEANS		1775.	1716.	1487.	3154.	2269.	2776.	2196.
SITE INDEX		1775.	1716.	1487.	3154.	2269.	2776.	2196.

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

ENVIRONMENT	MEAN	SE	DUNCAN GROUPS	LSD TESTS
RO 2006	3153.9	57.813	.	
NA 2006	2775.6	57.813	3.	
TY 2006	2269.4	57.813	33.	
VR 2006	1775.4	57.813	| 333.	
LA 2006	1716.0	57.813	| 333..	
ME 2006	1487.4	57.813	33332.	

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

TREATMENT		MEAN	SE	DUNCAN GROUPS	LSD TESTS
CA	CA	2395.5	105.55	| .	
Y3	Y3	2379.0	105.55	| ..	
YC	YC	2336.0	105.55	| ...	
CC	CC	2288.9	105.55	||	
Y1	Y1	2274.8	105.55	||	
YA	YA	2274.0	105.55	||	
YB	YB	2263.8	105.55	||	
Y2	Y2	2260.4	105.55	|||	
DC	DC	2236.5	105.55	|||	
DB	DB	2200.6	105.55	|||	
YD	YD	2200.0	105.55	|||	
DE	DE	2183.5	105.55	|||	
CE	CE	2173.9	105.55	|||	
DH	DH	2149.1	105.55	|||	
DF	DF	2136.8	105.55	|||	
DD	DD	2125.6	105.55	|||	
D1	D1	2124.0	105.55	|||	
CD	CD	2088.8	105.55	||| 1.....	
DG	DG	1945.0	105.55	|| 22111111.....	
CB	CB	1889.4	105.55	| 22221111111.....	

RESIDUALS FROM THE ADDITIVE TREATMENT BY SITE MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN STANDARD ERRORS, ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		R O	N A	T Y	V R	L A	M E	T - E
		2	2	2	2	2	2	F
		0	0	0	0	0	0	C
		0	0	0	0	0	0	T
		6	6	6	6	6	6	S
CA	CA	0	0	0	0	0	0	1
Y3	Y3	0	1	0	-1	0	0	3
YC	YC	0	0	0	0	-1	0	1
CC	CC	0	2	0	0	0	-1	0
Y1	Y1	0	0	0	-1	0	0	0
YA	YA	0	0	0	0	0	0	0
YB	YB	0	-4	0	2	0	0	0
Y2	Y2	0	1	1	-1	0	0	0
DC	DC	-1	0	0	0	1	0	0
DB	DB	0	0	0	0	0	0	0
YD	YD	-1	0	1	-1	1	0	0
DE	DE	0	0	0	0	0	0	0
CE	CE	0	0	0	0	0	1	0
DH	DH	1	0	0	-1	0	0	0
DF	DF	0	0	0	0	0	0	0
DD	DD	0	0	0	1	0	0	0
D1	D1	0	0	-1	0	-1	1	0
CD	CD	0	-1	-2	2	1	0	-1
DG	DG	0	-1	0	1	0	0	-2
CB	CB	0	0	0	1	-1	-1	-2
SITE EFFECTS		18	0	1	-7	-9	-13	93

```
BOX PLOT OF  120 STUDENTIZED RESIDUALS FROM LPLT= -2.590    TO ULPT=  2.632
NO.<LPLT                                                          NO.>UPLT
       1 *      ----------------I     +      I--------------    **  *   0

MEDIAN= -0.5276E-01 ANDERSON-DARLING STATISTIC=  0.548

ANALYSIS OF VARIANCE FOR THE ADDITIVE MODEL

-------------------------------------------------------------------
SOURCE                  D.F.     S.S.         M.S.     F       FPROB
-------------------------------------------------------------------
TREATMENTS               19   0.185535E+07    97650.2
LOCATIONS                 5   0.433662E+08   0.867323E+07
TREATMENT X SITES        95   0.635051E+07    66847.4
-------------------------------------------------------------------
TOTAL                   119   0.515720E+08
```

RESIDUALS FROM THE ADDITIVE SITE X TREATMENT MODEL FILE 06_ELITE   15/ 7/ 9 22:48

:PAGE 5

VARIETY\SITE		VR 2006	LA 2006	ME 2006	RO 2006	TY 2006	NA 2006	T-EFCTS
CA	CA	-188.2	101.7	-48.80	223.7	-217.6	129.2	199.2
Y3	Y3	-452.6	-35.49	206.0	-220.8	70.57	432.3	182.8 ***
YC	YC	183.3	-319.5	-220.0	195.1	224.1	-62.85	139.7
CC	CC	12.22	-209.9	-266.8	-18.53	-61.10	544.2 *	92.63
Y1	Y1	-442.8	122.3	-48.60	118.9	100.8	149.4	78.55
YA	YA	-120.7	130.2	-54.54	-42.19	11.08	76.17	77.73
YB	YB	605.6 **	102.3	-74.35	112.5	208.7	-954.7 ***	67.49
Y2	Y2	-255.0	-45.06	-57.63	-217.0	333.9	240.8	64.15
DC	DC	-5.743	431.1	92.22	-379.5	-66.88	-71.13	40.23
DB	DB	7.491	-229.0	92.58	208.2	-172.0	92.78	4.332
YD	YD	-263.9	253.1	-106.5	-262.4	265.3	114.4	3.711
DE	DE	-93.05	41.69	138.2	-61.09	31.74	-57.46	-12.76
CE	CE	-87.61	-89.13	231.4	126.7	-123.1	-58.32	-22.42
DH	DH	-307.5	-21.78	6.512	284.4	-85.90	124.2	-47.18
DF	DF	50.29	-90.27	126.4	-33.08	128.2	-181.6	-59.43
DD	DD	306.5	-87.46	110.5	-141.2	-125.6	-62.75	-70.72
D1	D1	-33.04	-259.9	290.5	65.01	-245.0	182.4	-72.30
CD	CD	518.4 *	398.5	-219.2	166.9	-595.9 *	-268.7	-107.5
DG	DG	241.9	63.61	36.89	-27.93	93.21	-407.7	-251.3 *
CB	CB	324.4	-256.8	-234.7	-97.53	225.4	39.26	-306.9 **
SITE EFFECTS		-420.9 ***	-480.3 ***	-708.9 ***	957.6 ***	73.11	579.3	2196. ***

FITTED VALUES FROM ADDITIVE SITE X TREATMENT MODEL FILE 06_ELITE   15/ 7/ 9 22:48

:PAGE 6

VARIETY\SITE		VR 2006	LA 2006	ME 2006	RO 2006	TY 2006	NA 2006	T-ESTS.
CA	CA	1975.	1915.	1687.	3353.	2469.	2975.	2395.
Y3	Y3	1958.	1899.	1670.	3337.	2452.	2958.	2379.
YC	YC	1915.	1856.	1627.	3294.	2409.	2915.	2336.
CC	CC	1868.	1809.	1580.	3247.	2362.	2868.	2289.
Y1	Y1	1854.	1795.	1566.	3232.	2348.	2854.	2275.
YA	YA	1853.	1794.	1565.	3232.	2347.	2853.	2274.
YB	YB	1843.	1783.	1555.	3221.	2337.	2843.	2264.
Y2	Y2	1840.	1780.	1552.	3218.	2334.	2840.	2260.
DC	DC	1816.	1756.	1528.	3194.	2310.	2816.	2237.
DB	DB	1780.	1720.	1492.	3158.	2274.	2780.	2201.
YD	YD	1779.	1720.	1491.	3158.	2273.	2779.	2200.
DE	DE	1763.	1703.	1475.	3141.	2257.	2763.	2184.
CE	CE	1753.	1694.	1465.	3131.	2247.	2753.	2174.
DH	DH	1728.	1669.	1440.	3107.	2222.	2728.	2149.
DF	DF	1716.	1657.	1428.	3094.	2210.	2716.	2137.
DD	DD	1705.	1645.	1417.	3083.	2199.	2705.	2126.
D1	D1	1703.	1644.	1415.	3082.	2197.	2703.	2124.
CD	CD	1668.	1609.	1380.	3046.	2162.	2668.	2089.
DG	DG	1524.	1465.	1236.	2903.	2018.	2524.	1945.
CB	CB	1468.	1409.	1181.	2847.	1963.	2469.	1889.
SITE ESTS.		1775.	1716.	1487.	3154.	2269.	2776.	2196.

 REGRESSIONS OF STY_KGHA FOR EACH VARIETY ON MEANS OF STY_KGHA AT EACH SITE

	VARIETY	MEAN	SLOPE	SE	MS-TXL	MS-REG	MS-DEV	R**2(%)
CA	CA	2395.48	1.156	0.112	32444.62	52731.46	27372.90	33.
CB	CB	1889.42	1.046	0.181	57625.39	4518.93	70902.00	2.
CC	CC	2288.91	1.267	0.174	83131.64	154025.53	65408.16	37.
CD	CD	2088.81	0.865	0.321	186136.75	39765.19	222729.64	4.
CE	CE	2173.86	0.997	0.109	20757.81	15.63	25943.36	0.
D1	D1	2123.98	1.038	0.169	50107.24	3162.52	61843.42	1.
DB	DB	2200.61	1.130	0.111	28520.82	36608.82	26498.82	26.
YA	YA	2274.01	1.015	0.069	8443.59	456.69	10440.32	1.
DC	DC	2236.51	0.687	0.125	69584.23	212948.50	33743.15	61.
YB	YB	2263.77	0.686	0.362	270081.06	214120.91	284071.09	16.
YC	YC	2335.96	1.184	0.153	55265.76	73450.07	50719.68	27.
DD	DD	2125.56	0.840	0.106	30690.15	55247.07	24550.92	36.
DE	DE	2183.52	0.922	0.053	7505.28	13057.99	6117.11	35.
DF	DF	2136.85	0.910	0.083	15429.94	17514.72	14908.75	23.
DG	DG	1944.96	0.809	0.136	47928.62	79303.84	40084.81	33.
DH	DH	2149.09	1.218	0.105	39744.52	103299.01	23855.90	52.
Y1	Y1	2274.83	1.171	0.151	52003.28	63089.71	49231.67	24.
Y2	Y2	2260.43	1.058	0.180	57387.31	7315.95	69905.16	3.
YD	YD	2199.99	0.954	0.184	59475.14	4670.78	73176.23	2.
Y3	Y3	2379.03	1.049	0.236	97838.30	5153.33	121009.55	1.

 SLOPE - SLOPES OF REGRESSIONS OF VARIETY MEANS ON SITE INDEX.
          * INDICATES SLOPES SIGNIFICANTLY DIFFERENT FROM THE
          SLOPE FOR THE OVERALL REGRESSION WHICH IS   1.00
 MS-TXL - CONTRIBUTION OF EACH VARIETY TO INTERACTION MS
 MS-REG - CONTRIBUTION OF EACH VARIETY TO THE REGRESSION
          COMPONENT OF THE TREATMENT BY LOCATION INTERACTION
 MS-DEV - DEVIATIONS FROM REGRESSION COMPONENT OF INTERACTION
 R**2   - SQUARED CORRELATION BETWEEN RESIDUALS FROM THE MAIN
          EFFECTS MODEL AND THE SITE INDEX.

```
VARIATE STY_KGHA WAS SITE INDEX WITH OVERALL MEAN  2196.
THE FOLLOWING SITE MEANS OF STY_KGHA WERE USED AS X-VARIATES
          1775.     1716.     1487.     3154.     2269.     2776.
-----------------------------------------------------------------
ANOVA FOR VARIABLE STY_KGHA WITH SITE REGRESSIONS ON STY_KGHA
-----------------------------------------------------------------
SOURCE             D.F.      S.S.         M.S.     F       FPROB
-----------------------------------------------------------------
TREATMENTS          19   0.185535E+07    97650.2
LOCATIONS            5   0.433662E+08   0.867323E+07
TREATMENT X SITES   95   0.635051E+07    66847.4
   TRT X SITE REG   19   0.114046E+07    60024.0       0.876   0.613
   DEVIATIONS       76   0.521005E+07    68553.3
-----------------------------------------------------------------
TOTAL              119   0.515720E+08
```

```
AMMI ANALYSIS  FILE 06_ELITE   15/ 7/ 9 22:48
 ------------------------------------------------------------- :PAGE   8
 SINGULAR VALUES OF INTERACTION MATRIX (CONDITION=  0)
 1757.0    1062.4    1011.3    795.26    692.44

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR TREATMENTS

 CA CA          CA          -0.394199E+01-0.840790E+01 0.439197E+01 0.147221E+01
 CB CB          CB           0.359027E+01 0.434907E+01-0.129164E+02-0.968057E+01
 CC CC          CC          -0.923469E+01-0.623241E+01-0.858281E+01-0.137225E+02
 CD CD          CD           0.157411E+02-0.156787E+02 0.114999E+02-0.108346E+02
 CE CE          CE          -0.566633E+00-0.466858E+01 0.507329E+00 0.962328E+01
 D1 D1          D1          -0.440872E+01-0.969749E+01-0.328971E+01 0.641099E+01
 DB DB          DB          -0.152152E+01-0.920101E+01-0.540939E+01 0.470031E+01
 YA YA          YA          -0.287915E+01 0.147092E+01 0.398659E+01-0.216485E+01
 DC DC          DC           0.135090E+01 0.635268E+01 0.155654E+02-0.545059E+01
 YB YB          YB           0.264529E+02 0.869518E+01-0.211254E+01 0.567669E+01
 YC YC          YC           0.363139E+01 0.112264E+01-0.148506E+02-0.101506E+01
 DD DD          DD           0.533627E+01-0.240754E+01-0.162022E+01-0.279437E+01
 DE DE          DE          -0.827122E+00 0.263872E+01 0.298322E+01 0.437064E+01
 DF DF          DF           0.315360E+01 0.437273E+01-0.254943E+01 0.555411E+01
 DG DG          DG           0.107660E+02 0.505095E+01 0.636019E+00 0.302878E+01
 DH DH          DH          -0.625791E+01-0.602856E+01 0.595441E+00 0.675100E+01
 Y1 Y1          Y1          -0.902759E+01 0.216971E+01 0.413435E+01 0.424361E+01
 Y2 Y2          Y2          -0.927467E+01 0.106775E+02-0.262935E+01-0.307413E+01
 YD YD          YD          -0.630322E+01 0.120044E+02 0.660841E+01-0.511646E+01
 Y3 Y3          Y3          -0.157792E+02 0.341764E+01 0.305183E+01 0.202148E+01

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR ENVIRONMENTS

 VR             VR 2006      0.277337E+02-0.506345E+01-0.882433E+01-0.121522E+02
 LA             LA 2006      0.483691E+01 0.439870E+01 0.263694E+02-0.611886E+01
 ME             ME 2006     -0.344280E+01-0.107758E+01 0.563889E+01 0.179359E+02
 RO             RO 2006      0.419757E+01-0.160050E+02-0.714860E+01 0.110433E+02
 TY             TY 2006     -0.288400E+01 0.262317E+02-0.116678E+02 0.203382E+01
 NA             NA 2006     -0.304414E+02-0.848438E+01-0.436758E+01-0.127419E+02
```

RESIDUALS FROM THE AMMI-2 MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN UNITS OF ROOT (RESIDUAL GXE MS), ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		R O 2 0 0 6	N A 2 0 0 6	T Y 2 0 0 6	V R 2 0 0 6	L A 2 0 0 6	M E 2 0 0 6	T - E F C T S
CA	CA	0	0	0	0	0	0	
Y3	Y3	0	0	0	0	0	0	
YC	YC	0	0	1	0	-1	-1	
CC	CC	0	1	0	1	0	-1	
Y1	Y1	0	0	0	0	0	0	
YA	YA	0	0	0	0	0	0	
YB	YB	0	0	0	0	0	0	
Y2	Y2	0	0	0	0	0	0	
DC	DC	-1	0	-1	0	1	0	
DB	DB	0	0	0	0	0	0	
YD	YD	0	0	0	0	1	0	
DE	DE	0	0	0	0	0	0	
CE	CE	0	0	0	0	0	1	
DH	DH	1	0	0	0	0	0	
DF	DF	0	0	0	0	0	0	
DD	DD	0	0	0	0	0	0	
D1	D1	0	0	0	0	0	1	
CD	CD	0	0	0	0	1	0	
DG	DG	0	0	0	0	0	0	
CB	CB	0	0	0	1	-1	-1	

```
BOX PLOT OF  120 STANDERSIZED RESIDUALS FROM LPLT= -1.672    TO ULPT=  1.938
NO.<LPLT                                                          NO.>UPLT
       0 *  ------------------I     +      I--------------          *   0
```

ANALYSIS OF VARIANCE FOR THE AMMI MODEL

SOURCE	D.F.	S.S.	M.S.	F	FPROB
TREATMENTS	19	0.185535E+07	97650.2		
LOCATIONS	5	0.433662E+08	0.867323E+07		
TREATMENT X SITES	95	0.635051E+07	66847.4		
AMMI COMPONENT 1	23	0.308713E+07	134223.	2.961	0.000
AMMI COMPONENT 2	21	0.112868E+07	53746.6	1.284	0.230
AMMI COMPONENT 3	19	0.102278E+07	53830.6	1.549	0.134
AMMI COMPONENT 4	17	632439.	37202.3	1.164	0.387
GXE RESIDUAL	15	479473.			
TOTAL	119	0.515720E+08			

 GENOTYPE MAP SHOWING BEST GENOTYPES OVER THE RANGE OF AMMI-2 SITE SCORES (2 CHRS/PIXEL)

```
 26.23 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 25.43 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 24.64 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y2Y2Y2Y2Y2Y2Y2Y2Y2YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 23.84 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y2Y2Y2Y2Y2Y2Y2Y2YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 23.04 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y2Y2Y2Y2Y2Y2Y2Y2YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 22.25 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y2Y2Y2Y2Y2Y2Y2Y2YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 21.45 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y2Y2Y2Y2Y2Y2Y2Y2YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 20.65 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y2Y2Y2Y2Y2Y2Y2Y2YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 19.86 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y2Y2Y2Y2Y2Y2Y2Y2YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 19.06 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y2Y2Y2Y2Y2Y2Y2Y2YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 18.26 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y2Y2Y2YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 17.47 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y2Y2YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 16.67 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y2YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 15.87 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 15.07 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 14.28 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 13.48 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 12.68 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 11.89 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 11.09 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 10.29 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
  9.50 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
  8.70 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
  7.90 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
  7.11 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
  6.31 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
  5.51 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
  4.71 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
  3.92 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3YBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
  3.12 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
  2.32 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
  1.53 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
  0.73 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 -0.07 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 -0.86 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 -1.66 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
```

```
 -2.46 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 -3.25 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 -4.05 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 -4.85 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 -5.65 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 -6.44 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 -7.24 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 -8.04 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 -8.83 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
 -9.63 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
-10.43 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACACACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
-11.22 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACACACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
-12.02 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACACACACACACACACACAYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
-12.82 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACACACACACACACACACACDCDYBYBYBYBYBYBYBYBYBYBYBYBYBYBYBYB
-13.61 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACACACACACACACACACACACDCDCDCDYBYBYBYBYBYBYBYBYBYBYBYBYB
-14.41 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACACACACACACACACACACACACDCDCDCDCDYBYBYBYBYBYBYBYBYBYBYBYB
-15.21 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACACACACACACACACACACACACACDCDCDCDCDCDCDCDYBYBYBYBYBYBYBYBYB
-16.00 Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3Y3CACACACACACACACACACACACACACACACACACACACACACACACACACACDCDCDCDCDCDCDCDCDCDYBYBYBYBYBYBYB
        ^           ^           ^           ^           ^           ^           ^           ^           ^
    -0.304E+02  -0.239E+02  -0.173E+02  -0.107E+02  -0.410E+01   0.249E+01   0.907E+01   0.157E+02   0.222E+02
```

AMMI RESIDUALS, ADDITIVE EFFECTS AND MULTIPLICATIVE SCORES FILE 06_ELITE   15/ 7/ 9 22:48
 -- :PAGE 10

 SECTION 1

VARIETY\SITE		VR 2006		LA 2006		ME 2006		RO 2006		TY 2006		NA 2006		T-EFCTS	
CA	CA	-121.4		157.7		-71.43		105.7		-8.430		-62.11		199.2	***
Y3	Y3	2.334		25.80		155.3		-99.89		-64.59		-19.00		182.8	***
YC	YC	88.24		-342.1		-206.3		197.8		205.1		57.22		139.7	
CC	CC	236.8		-137.9		-305.4		-79.52		75.75		210.2		92.63	***
Y1	Y1	-181.4		156.5		-77.34		191.5		17.81		-107.0		78.55	
YA	YA	-33.45		137.7		-62.86		-6.566		-35.81		1.001		77.73	
YB	YB	-84.06		-63.94		26.09		140.7		56.92		-75.69		67.49	***
Y2	Y2	56.28		-47.16		-78.06		-7.175		27.06		49.06		64.15	
DC	DC	-11.04		396.6		103.7		-283.5		-229.6		23.89		40.23	
DB	DB	3.099		-181.2		77.43		67.29		65.00		-31.61		4.332	
YD	YD	-28.26		230.8		-115.3		-43.84		-67.77		24.34		3.711	***
DE	DE	-56.75		34.09		138.2		-15.38		-39.87		-60.25		-12.76	***
CE	CE	-95.53		-65.85		224.5		54.36		-2.261		-115.2		-22.42	
DH	DH	-164.4		35.01		-21.53		214.1		54.20		-117.4		-47.18	
DF	DF	-15.03		-124.8		142.0		23.67		22.62		-48.45		-59.43	***
DD	DD	146.3		-102.7		126.3		-202.1		-47.05		79.27		-70.72	***
D1	D1	40.13		-195.9		264.9		-71.69		-3.292		-34.11		-72.30	***
CD	CD	2.466		391.3		-181.9		-150.2		-139.2		77.48		-107.5	
DG	DG	-31.06		-10.68		79.40		7.724		-8.241		-37.13		-251.3	***
CB	CB	246.9		-293.3		-217.6		-42.99		121.7		185.4		-306.9	
SITE EFFECTS		-420.9	***	-480.3	***	-708.9	***	957.6		73.11	***	579.3		2196.	
AMMI1 SITE		27.73	***	4.837	***	-3.443	***	4.198	***	-2.884		-30.44	***	.	
AMMI2 SITE		-5.063		4.399	***	-1.078	***	-16.00	***	26.23		-8.484		.	

SECTION 2

VARIETY\SITE		AMMI1 TRT		AMMI2 TRT	
CA	CA	-3.942	***	-8.408	***
Y3	Y3	-15.78		3.418	***
YC	YC	3.631		1.123	
CC	CC	-9.235	***	-6.232	***
Y1	Y1	-9.028	***	2.170	***
YA	YA	-2.879		1.471	***
YB	YB	26.45	***	8.695	
Y2	Y2	-9.275		10.68	***
DC	DC	1.351		6.353	***
DB	DB	-1.522		-9.201	***
YD	YD	-6.303		12.00	
DE	DE	-.8271		2.639	
CE	CE	-.5666	***	-4.669	***
DH	DH	-6.258	***	-6.029	***
DF	DF	3.154		4.373	
DD	DD	5.336		-2.408	
D1	D1	-4.409	***	-9.697	
CD	CD	15.74	***	-15.68	
DG	DG	10.77	***	5.051	
CB	CB	3.590	***	4.349	
SITE EFFECTS		.		.	
AMMI1 SITE		.		.	
AMMI2 SITE		.		.	

FITTED VALUES FROM THE AMMI MODEL FILE 06_ELITE   15/ 7/ 9 22:48
 -- :PAGE 11

 SECTION 1

VARIETY\SITE		VR 2006	LA 2006	ME 2006	RO 2006	TY 2006	NA 2006	T-ESTS.
CA	CA	1908.	1859.	1709.	3471.	2259.	3166.	2395.
Y3	Y3	1503.	1837.	1721.	3216.	2587.	3410.	2379.
YC	YC	2010.	1878.	1613.	3291.	2428.	2795.	2336.
CC	CC	1643.	1737.	1619.	3307.	2225.	3202.	2289.
Y1	Y1	1593.	1760.	1595.	3160.	2431.	3111.	2275.
YA	YA	1766.	1786.	1573.	3196.	2394.	2929.	2274.
YB	YB	2532.	1950.	1454.	3193.	2489.	1964.	2264.
Y2	Y2	1528.	1782.	1572.	3008.	2640.	3032.	2260.
DC	DC	1821.	1791.	1516.	3098.	2472.	2721.	2237.
DB	DB	1784.	1672.	1507.	3299.	2037.	2904.	2201.
YD	YD	1543.	1742.	1500.	2939.	2606.	2869.	2200.
DE	DE	1726.	1711.	1475.	3095.	2328.	2766.	2184.
CE	CE	1761.	1670.	1472.	3204.	2126.	2810.	2174.
DH	DH	1585.	1612.	1468.	3177.	2082.	2970.	2149.
DF	DF	1781.	1691.	1412.	3038.	2316.	2583.	2137.
DD	DD	1865.	1660.	1401.	3144.	2120.	2563.	2126.
D1	D1	1630.	1580.	1441.	3218.	1955.	2920.	2124.
CD	CD	2184.	1616.	1343.	3363.	1705.	2322.	2089.
DG	DG	1797.	1539.	1194.	2867.	2120.	2154.	1945.
CB	CB	1546.	1446.	1164.	2792.	2066.	2323.	1889.
SITE ESTS.		1775.	1716.	1487.	3154.	2269.	2776.	2196.
AMMI1 SITE		27.73	4.837	-3.443	4.198	-2.884	-30.44	.
AMMI2 SITE		-5.063 I	4.399 I	-1.078 I	-16.00 I	26.23 I	-8.484 I	.

SECTION 2

VARIETY\SITE		AMMI1 TRT	AMMI2 TRT
CA	CA	-3.942	-8.408
Y3	Y3	-15.78	3.418
YC	YC	3.631	1.123
CC	CC	-9.235	-6.232
Y1	Y1	-9.028	2.170
YA	YA	-2.879	1.471
YB	YB	26.45	8.695
Y2	Y2	-9.275	10.68
DC	DC	1.351	6.353
DB	DB	-1.522	-9.201
YD	YD	-6.303	12.00
DE	DE	-.8271	2.639
CE	CE	-.5666	-4.669
DH	DH	-6.258	-6.029
DF	DF	3.154	4.373
DD	DD	5.336	-2.408
D1	D1	-4.409	-9.697
CD	CD	15.74	-15.68
DG	DG	10.77	5.051
CB	CB	3.590	4.349
SITE ESTS.		.	.
AMMI1 SITE		.	.
AMMI2 SITE		.	.

Addendum 7: Cross-site analysis of total starch yield, the 2007 season trials

```
PBGXE - CROSS SITE ANALYSIS  FILE 07_ELITE   16/ 7/ 9  0:33
 ------------------------------------------------------------------ :PAGE   1

  20       CN$    CODES:
  1 CA          CA            2 CC          CC            3 CE          CE
  4 CD          CD            5 D1          D1            6 D2          D2
  7 D3          D3            8 D4          D4            9 YC          YC
 10 Y2          Y2           11 EA          EA           12 EB          EB
 13 G1          G1           14 G2          G2           15 H1          H1
 16 H2          H2           17 H3          H3           18 H4          H4
 19 H5          H5           20 H6          H6

   9     SITE$    CODES:
  1 PI 2007       2 KL 2007       3 LA 2007       4 ME 2007       5 RO 2007
  6 TY 2007       7 NA 2007       8 RI 2007       9 AL 2007

 TREATMENT BY VARIATE MEANS HAVE BEEN REQUESTED FOR  1 VARIATES:
 STY_KGHA
 GXE ANALYSIS HAS BEEN REQUESTED FOR  1 VARIATES
```

TREATMENT MEANS AND COUNTS OVER SITES FOR EACH VARIATE. FILE 07_ELITE 16/ 7/ 9 0:33
:PAGE 2

VARIETY\SITE		STY_KGHA	
CA	CA	2977.	27
CC	CC	3036.	27
CE	CE	2927.	27
CD	CD	2194.	27
D1	D1	3216.	27
D2	D2	3283.	27
D3	D3	3161.	27
D4	D4	3263.	27
YC	YC	3122.	27
Y2	Y2	3066.	27
EA	EA	3132.	27
EB	EB	2657.	27
G1	G1	3267.	27
G2	G2	3364.	27
H1	H1	3148.	27
H2	H2	3148.	27
H3	H3	3254.	27
H4	H4	2954.	27
H5	H5	2583.	27
H6	H6	3193.	27
SITE MEANS		3047.	540

20 X 9 MATRIX OF TREATMENT BY SITE MEANS FOR VARIATE STY_KGHA FILE 07_ELITE 16/ 7/ 9 0:33
:PAGE 3

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
CA	CA	3117.	2662.	3511.	1972.	4089.	3123.	3447.
CC	CC	2601.	2442.	3161.	2094.	3868.	2989.	3425.
CE	CE	2964.	2659.	2779.	2207.	3519.	2919.	2885.
CD	CD	3073.	3121.	2675.	1094.	3754.	2091.	362.7
D1	D1	3122.	2721.	3561.	2536.	3941.	3406.	3307.
D2	D2	2901.	2910.	3391.	2423.	4176.	3093.	3463.
D3	D3	2948.	2392.	3148.	2039.	4363.	3250.	3727.
D4	D4	2803.	2949.	3275.	2035.	4498.	3247.	3726.
YC	YC	3091.	2762.	3140.	1702.	4118.	3390.	3241.
Y2	Y2	2876.	2518.	3537.	2327.	3277.	3347.	3640.
EA	EA	2752.	2657.	3811.	2566.	3466.	3305.	3124.
EB	EB	2301.	2453.	3049.	1869.	3197.	2671.	2680.
G1	G1	3145.	2482.	3732.	2006.	4456.	3222.	3462.
G2	G2	3429.	2232.	3581.	2193.	4602.	3526.	3488.
H1	H1	3118.	2462.	3683.	2151.	3385.	3034.	3638.
H2	H2	3207.	2836.	3242.	1891.	3687.	3077.	3781.
H3	H3	3027.	2803.	3605.	2410.	3757.	3690.	3208.
H4	H4	2883.	2865.	3239.	1644.	3239.	3193.	2910.
H5	H5	2985.	2370.	2827.	1762.	3643.	2158.	2146.
H6	H6	3049.	2550.	3225.	1964.	3988.	3588.	3851.
SITE MEANS		2970.	2642.	3309.	2044.	3851.	3116.	3176.
SITE INDEX		2970.	2642.	3309.	2044.	3851.	3116.	3176.

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	TRT MEANS
CA	CA	1961.	2912.	2977.
CC	CC	3046.	3702.	3036.
CE	CE	3305.	3106.	2927.
CD	CD	1095.	2479.	2194.
D1	D1	2857.	3490.	3216.
D2	D2	3533.	3655.	3283.
D3	D3	3227.	3353.	3161.
D4	D4	3161.	3674.	3263.
YC	YC	3083.	3570.	3122.
Y2	Y2	2566.	3509.	3066.
EA	EA	2912.	3597.	3132.
EB	EB	2661.	3036.	2657.
G1	G1	3099.	3798.	3267.
G2	G2	3233.	3996.	3364.
H1	H1	3022.	3838.	3148.
H2	H2	3058.	3558.	3148.
H3	H3	3141.	3646.	3254.
H4	H4	3350.	3259.	2954.
H5	H5	2456.	2904.	2583.
H6	H6	3145.	3378.	3193.
SITE MEANS		2896.	3423.	3047.
SITE INDEX		2896.	3423.	3047.

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

ENVIRONMENT	MEAN	SE	DUNCAN GROUPS	LSD TESTS
RO 2007	3851.1	80.388		.
AL 2007	3423.0	80.388	|	3.
LA 2007	3308.6	80.388	||	3..
NA 2007	3175.5	80.388	||	31..
TY 2007	3115.8	80.388	|||	32...
PI 2007	2969.6	80.388	||	332...
RI 2007	2895.7	80.388	|	3331...
KL 2007	2642.2	80.388		3333321.
ME 2007	2044.3	80.388		33333333.

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

TREATMENT		MEAN	SE	DUNCAN GROUPS	LSD TESTS
G2	G2	3364.5	119.84	|	.
D2	D2	3282.9	119.84	||	..
G1	G1	3266.8	119.84	||	...
D4	D4	3263.2	119.84	||	....
H3	H3	3254.1	119.84	||	.....
D1	D1	3215.6	119.84	||	.....
H6	H6	3193.2	119.84	||	.....
D3	D3	3160.8	119.84	||	.....
H2	H2	3148.4	119.84	||	.....
H1	H1	3147.9	119.84	||	.....
EA	EA	3132.2	119.84	||	.....
YC	YC	3121.8	119.84	||	.....
Y2	Y2	3066.3	119.84	||	.....
CC	CC	3036.3	119.84	||	.....
CA	CA	2977.1	119.84	|||	1.....
H4	H4	2953.6	119.84	||	1.....
CE	CE	2926.9	119.84	|||	1111.....
EB	EB	2657.4	119.84	||	33333222222211....
H5	H5	2583.5	119.84	|	33333333222222111..
CD	CD	2193.8	119.84		3333333333333333321.

RESIDUALS FROM THE ADDITIVE TREATMENT BY SITE MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN STANDARD ERRORS, ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		ROAL	ANTY	PIRI	RILE	MET-	
		2 2 2 2 2 2 2 2 2	0 0 0 0 0 0 0 0 0	0 0 0 0 0 0 0 0 0	7 7 7 7 7 7 7 7 7		TSCF
G2	G2	1 0 0 0 0 0 0 -2 0					2
D2	D2	0 0 0 0 0 0 1 0 0					2
G1	G1	1 0 0 0 0 0 0 -1 0					1
D4	D4	1 0 0 1 0 -1 0 0 0					1
H3	H3	0 0 0 0 1 0 0 0 0					1
D1	D1	0 0 0 0 0 0 0 0 0					1
H6	H6	0 0 0 1 0 0 0 0 0					1
D3	D3	1 0 0 1 0 0 0 -1 0					0
H2	H2	0 0 0 1 0 0 0 0 0					0
H1	H1	-1 0 0 1 0 0 0 0 0					0
EA	EA	-1 0 1 0 0 0 0 0 1					0
YC	YC	0 0 0 0 0 0 0 0 -1					0
Y2	Y2	-1 0 0 1 0 0 -1 0 0					0
CC	CC	0 0 0 0 0 -1 0 0 0					0
CA	CA	0 -1 0 1 0 0 -2 0 0					0
H4	H4	-1 0 0 0 0 0 1 0 0					0
CE	CE	0 0 -1 0 0 0 1 0 0					-1
EB	EB	0 0 0 0 0 0 0 0 0					-3
H5	H5	0 0 0 -1 -1 1 0 0 0					-3
CD	CD	2 0 0 -5 0 2 -2 4 0					-7
SITE EFFECTS		10 0 3 1 0 -1 -2 -5 -13					113

```
BOX PLOT OF  180 STUDENTIZED RESIDUALS FROM LPLT= -2.866    TO ULPT=  2.895
NO.<LPLT                                                          NO.>UPLT
       1 *  *   *   -------------I     +    I-----------      *     *   1

MEDIAN=  0.5603E-01 ANDERSON-DARLING STATISTIC=  1.935 **

ANALYSIS OF VARIANCE FOR THE ADDITIVE MODEL

-------------------------------------------------------------------
SOURCE                  D.F.     S.S.         M.S.     F       FPROB
-------------------------------------------------------------------
TREATMENTS               19   0.136225E+08    716976.
LOCATIONS                 8   0.415143E+08   0.518928E+07
TREATMENT X SITES       152   0.196454E+08    129246.
-----------------------------------------------------------------
TOTAL                   179   0.747822E+08
```

RESIDUALS FROM THE ADDITIVE SITE X TREATMENT MODEL FILE 07_ELITE   16/ 7/ 9 0:33
 -- :PAGE   5

 SECTION 1

VARIETY\SITE		PI 2007		KL 2007		LA 2007		ME 2007		RO 2007		TY 2007		NA 2007	
CA	CA	217.6		90.16		272.9		-1.902		307.7		77.84		341.6	
CC	CC	-357.5		-189.5		-136.9		60.93		28.13		-116.2		260.3	
CE	CE	114.7		137.0		-409.6		283.2		-211.6		-76.92		-169.8	
CD	CD	956.5	**	1332.	***	220.2		-96.35		756.3	*	-171.5		-1959.	***
D1	D1	-15.94		-89.56		84.17		323.1		-78.09		121.7		-37.22	
D2	D2	-304.0		32.16		-153.0		143.1		89.10		-258.8		52.29	
D3	D3	-135.1		-363.4		-274.1		-119.0		398.3		20.84		437.6	
D4	D4	-382.1		90.45		-249.1		-224.9		431.3		-85.12		334.7	
YC	YC	46.94		44.81		-243.3		-416.6		192.5		199.2		-9.190	
Y2	Y2	-112.2		-143.5		209.1		263.4		-593.2		212.1		445.2	
EA	EA	-303.0		-69.76		417.3		436.9		-470.3		104.5		-136.8	
EB	EB	-279.0		200.8		130.8		214.8		-264.3		-54.93		-106.0	
G1	G1	-44.38		-379.9		203.7		-257.8		385.3		-113.2		67.17	
G2	G2	142.3		-727.7	*	-44.84		-168.7		434.1		92.58		-4.572	
H1	H1	47.72		-280.9		273.6		6.138		-566.4		-182.0		361.6	
H2	H2	136.3		92.89		-167.8		-254.8		-265.6		-140.3		504.4	
H3	H3	-149.0		-46.33		89.98		159.1		-301.2		367.1		-174.3	
H4	H4	7.308		316.7		23.88		-306.3		-518.7		171.2		-171.6	
H5	H5	479.1		191.8		-17.36		181.8		255.9		-493.9		-566.2	
H6	H6	-66.19		-238.2		-229.7		-226.0		-9.176		325.9		530.1	
SITE EFFECTS		-77.69		-405.1	***	261.3	***	-1003.	***	803.8	***	68.54		128.2	

SECTION 2

VARIETY\SITE		RI 2007		AL 2007	T-EFCTS	
CA	CA	-864.7	**	-441.2	-70.20	
CC	CC	160.8		289.8	-10.97	
CE	CE	529.9		-196.8	-120.4	
CD	CD	-947.0	**	-90.76	-853.5	***
D1	D1	-207.2		-101.0	168.3	
D2	D2	402.2		-3.145	235.6	*
D3	D3	218.1		-183.2	113.5	
D4	D4	49.70		35.04	215.9	
YC	YC	113.3		72.39	74.53	
Y2	Y2	-348.3		67.37	18.94	
EA	EA	-68.26		89.45	84.91	
EB	EB	155.2		2.685	-389.9	**
G1	G1	-16.46		155.4	219.5	
G2	G2	20.61		256.2	317.2	**
H1	H1	26.23		314.0	100.5	
H2	H2	61.15		33.82	101.1	
H3	H3	38.27		16.40	206.8	
H4	H4	548.2		-70.56	-93.75	
H5	H5	24.15		-55.39	-463.8	***
H6	H6	103.9		-190.7	145.9	
SITE EFFECTS		-151.7	*	375.7	3047.	***

FITTED VALUES FROM ADDITIVE SITE X TREATMENT MODEL FILE 07_ELITE   16/ 7/ 9 0:33
 ---------- :PAGE   6

 SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
CA	CA	2899.	2572.	3238.	1974.	3781.	3046.	3105.
CC	CC	2959.	2631.	3298.	2033.	3840.	3105.	3165.
CE	CE	2849.	2522.	3188.	1924.	3731.	2995.	3055.
CD	CD	2116.	1789.	2455.	1191.	2998.	2262.	2322.
D1	D1	3138.	2810.	3477.	2213.	4019.	3284.	3344.
D2	D2	3205.	2878.	3544.	2280.	4087.	3351.	3411.
D3	D3	3083.	2756.	3422.	2158.	3965.	3229.	3289.
D4	D4	3185.	2858.	3524.	2260.	4067.	3332.	3391.
YC	YC	3044.	2717.	3383.	2119.	3926.	3190.	3250.
Y2	Y2	2989.	2661.	3328.	2063.	3870.	3135.	3194.
EA	EA	3055.	2727.	3393.	2129.	3936.	3201.	3260.
EB	EB	2580.	2252.	2919.	1654.	3461.	2726.	2786.
G1	G1	3189.	2862.	3528.	2264.	4071.	3335.	3395.
G2	G2	3287.	2959.	3626.	2362.	4168.	3433.	3493.
H1	H1	3070.	2743.	3409.	2145.	3952.	3216.	3276.
H2	H2	3071.	2743.	3410.	2145.	3952.	3217.	3277.
H3	H3	3176.	2849.	3515.	2251.	4058.	3323.	3382.
H4	H4	2876.	2548.	3215.	1951.	3757.	3022.	3082.
H5	H5	2506.	2178.	2845.	1581.	3387.	2652.	2712.
H6	H6	3116.	2788.	3454.	2190.	3997.	3262.	3321.
SITE ESTS.		2970.	2642.	3309.	2044.	3851.	3116.	3176.

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	T-ESTS.
CA	CA	2825.	3353.	2977.
CC	CC	2885.	3412.	3036.
CE	CE	2775.	3303.	2927.
CD	CD	2042.	2569.	2194.
D1	D1	3064.	3591.	3216.
D2	D2	3131.	3659.	3283.
D3	D3	3009.	3536.	3161.
D4	D4	3112.	3639.	3263.
YC	YC	2970.	3498.	3122.
Y2	Y2	2915.	3442.	3066.
EA	EA	2981.	3508.	3132.
EB	EB	2506.	3033.	2657.
G1	G1	3115.	3643.	3267.
G2	G2	3213.	3740.	3364.
H1	H1	2996.	3524.	3148.
H2	H2	2997.	3524.	3148.
H3	H3	3102.	3630.	3254.
H4	H4	2802.	3329.	2954.
H5	H5	2432.	2959.	2583.
H6	H6	3042.	3569.	3193.
SITE ESTS.		2896.	3423.	3047.

 REGRESSIONS OF STY_KGHA FOR EACH VARIETY ON MEANS OF STY_KGHA AT EACH SITE

VARIETY		MEAN	SLOPE	SE	MS-TXL	MS-REG	MS-DEV	R**2(%)
CA	CA	2977.11	1.136	0.293	161226.00	38211.24	178799.53	3.
CC	CC	3036.34	1.068	0.159	47252.07	9464.36	52650.32	3.
CE	CE	2926.89	0.611*	0.159	84862.48	313621.59	52182.61	46.
CD	CD	2193.77	0.997	0.746	101460.25	13.56	115595	2.62 0.
D1	D1	3215.58	0.841	0.099	24393.19	52548.72	20370.97	27.
D2	D2	3282.91	0.916	0.158	47101.36	14675.77	51733.59	4.
D3	D3	3160.77	1.232	0.196	83918.92	111611.18	79962.88	17.
D4	D4	3263.19	1.262	0.173	71971.79	141998.22	61968.01	25.
YC	YC	3121.84	1.246	0.119	41481.34	125160.84	29527.13	38.
Y2	Y2	3066.25	0.774	0.229	108399.51	106300.04	108699.43	12.
EA	EA	3132.22	0.700	0.193	90646.12	186254.67	76987.77	26.
EB	EB	2657.40	0.762	0.109	36203.27	117215.16	24630.14	40.
G1	G1	3266.81	1.405*	0.085	55560.06	340394.88	14869.37	77.
G2	G2	3364.49	1.428*	0.178	105416.44	380777.62	66079.12	45.
H1	H1	3147.85	0.936	0.224	92512.60	8388.01	104530.40	1.
H2	H2	3148.39	1.004	0.180	58738.46	35.20	67124.64	0.
H3	H3	3254.14	0.834	0.133	39423.01	57214.26	36881.41	18.
H4	H4	2953.56	0.830	0.230	103515.43	59774.32	109764.16	7.
H5	H5	2583.50	0.891	0.250	116665.23	24841.67	129782.88	3.
H6	H6	3193.20	1.127	0.197	74925.29	33524.35	80839.71	6.

 SLOPE - SLOPES OF REGRESSIONS OF VARIETY MEANS ON SITE INDEX.
          * INDICATES SLOPES SIGNIFICANTLY DIFFERENT FROM THE
          SLOPE FOR THE OVERALL REGRESSION WHICH IS   1.00
 MS-TXL - CONTRIBUTION OF EACH VARIETY TO INTERACTION MS
 MS-REG - CONTRIBUTION OF EACH VARIETY TO THE REGRESSION
          COMPONENT OF THE TREATMENT BY LOCATION INTERACTION
 MS-DEV - DEVIATIONS FROM REGRESSION COMPONENT OF INTERACTION
 R**2   - SQUARED CORRELATION BETWEEN RESIDUALS FROM THE MAIN
          EFFECTS MODEL AND THE SITE INDEX.

```
VARIATE STY_KGHA WAS SITE INDEX WITH OVERALL MEAN  3047.
THE FOLLOWING SITE MEANS OF STY_KGHA WERE USED AS X-VARIATES
          2970.     2642.     3309.     2044.     3851.     3116.     3176.     2896.     3423.
-----------------------------------------------------------------------
ANOVA FOR VARIABLE STY_KGHA WITH SITE REGRESSIONS ON STY_KGHA
-----------------------------------------------------------------------
SOURCE              D.F.      S.S.         M.S.     F       FPROB
-----------------------------------------------------------------------
TREATMENTS          19   0.136225E+08    716976.
LOCATIONS            8   0.415143E+08   0.518928E+07
TREATMENT X SITES  152   0.196454E+08    129246.
   TRT X SITE REG   19   0.212203E+07    111686.       0.848   0.647
   DEVIATIONS      133   0.175234E+08    131755.
-----------------------------------------------------------------------
TOTAL              179   0.747822E+08
```

```
AMMI ANALYSIS  FILE 07_ELITE   16/ 7/ 9  0:33
 ------------------------------------------------------------ :PAGE   8
 SINGULAR VALUES OF INTERACTION MATRIX (CONDITION=  0)
 3203.6    1761.1    1584.8    1129.2    945.64    868.42    790.80    469.40

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR TREATMENTS

 CA CA          CA          -0.476368E+01 0.920063E+01-0.232817E+02 0.103482E+02
 CC CC          CC           0.800775E+01 0.187924E+01 0.340890E+01-0.637650E+01
 CE CE          CE           0.239861E+00-0.707517E+01 0.153900E+02 0.296834E+01
 CD CD          CD          -0.501562E+02 0.733612E+00-0.116151E+01 0.179970E+01
 D1 D1          D1          -0.445155E+00-0.398911E+01-0.711331E+01-0.309380E+01
 D2 D2          D2           0.383220E+01-0.195293E+00 0.107919E+02-0.374577E+01
 D3 D3          D3           0.868445E+01 0.141026E+02 0.364781E+01 0.102733E+01
 D4 D4          D4           0.425447E+01 0.120508E+02 0.421518E+01 0.455495E+01
 YC YC          YC          -0.302133E+00 0.793277E+01 0.609841E+01 0.653908E+01
 Y2 Y2          Y2           0.834360E+01-0.112107E+02-0.156615E+02 0.267494E+01
 EA EA          EA           0.223408E+01-0.163900E+02-0.733897E+01-0.926201E+01
 EB EB          EB           0.609536E+00-0.106127E+02 0.281246E+01-0.128935E+01
 G1 G1          G1           0.193903E+01 0.111811E+02-0.221544E+01-0.996284E+01
 G2 G2          G2           0.346850E+01 0.144210E+02-0.100691E+01-0.152596E+02
 H1 H1          H1           0.890134E+01-0.890178E+01-0.597782E+01-0.493443E+01
 H2 H2          H2           0.629184E+01 0.584699E+00 0.678632E+00 0.118249E+02
 H3 H3          H3           0.123039E+01-0.935302E+01-0.155168E+01-0.141583E+01
 H4 H4          H4           0.883718E+00-0.120423E+02 0.126755E+02 0.108716E+02
 H5 H5          H5          -0.133619E+02 0.857946E+00 0.665358E+01-0.775538E+01
 H6 H6          H6           0.101082E+02 0.682551E+01-0.106344E+01 0.104864E+02

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR ENVIRONMENTS

 PI             PI 2007     -0.197146E+02 0.389920E+01-0.257177E+01 0.407679E+01
 KL             KL 2007     -0.257503E+02-0.109332E+02 0.655294E+01 0.197783E+02
 LA             LA 2007     -0.471659E+01-0.102075E+02-0.170340E+02-0.895770E+01
 ME             ME 2007     -0.564137E-01-0.159469E+02-0.557163E+01-0.124018E+02
 RO             RO 2007     -0.151620E+02 0.337365E+02 0.269821E+01-0.683229E+01
 TY             TY 2007      0.501315E+01-0.242194E+01-0.696958E+01 0.863696E+01
 NA             NA 2007      0.394987E+02 0.944423E+01-0.114130E+02 0.125611E+02
 RI             RI 2007      0.173830E+02-0.551593E+01 0.319770E+02-0.268161E+01
 AL             AL 2007      0.350510E+01-0.205441E+01 0.233184E+01-0.141797E+02
```

RESIDUALS FROM THE AMMI-2 MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN UNITS OF ROOT (RESIDUAL GXE MS), ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		ROW 2	ALA 2	NAY 2	TYPI 2	RIL 2	KLM 2	TEFCTS
G2	G2	0	1	0	-1	0	0	EA
D2	D2	0	0	0	0	-1	0	EA
G1	G1	0	0	1	0	0	0	EA
D4	D4	0	0	0	0	0	-1	EA
H3	H3	0	0	0	0	1	0	EA
D1	D1	0	0	0	0	0	0	EA
H6	H6	0	0	0	0	1	0	EA
D3	D3	0	0	0	0	0	0	EA
H2	H2	0	0	0	1	0	1	EA
H1	H1	0	1	0	0	-1	1	EA
EA	EA	0	0	1	0	0	0	EA
YC	YC	0	0	0	0	0	0	EA
Y2	Y2	0	0	0	0	0	0	EA
CC	CC	0	1	0	0	0	0	EA
CA	CA	0	-1	1	1	0	0	EA
H4	H4	0	0	0	0	0	0	EA
CE	CE	0	0	-1	0	0	0	EA
EB	EB	0	0	0	0	0	0	EA
H5	H5	0	0	0	0	-1	0	EA
CD	CD	0	0	0	0	0	0	EA
G2	G2	0	1	0	-1	0	0	EA
D2	D2	0	0	0	0	-1	0	EA
G1	G1	0	0	1	0	0	0	EA
D4	D4	0	0	0	0	0	-1	EA
H3	H3	0	0	0	0	1	0	EA
D1	D1	0	0	0	0	0	0	EA
H6	H6	0	0	0	0	1	0	EA
D3	D3	0	0	0	0	0	0	EA
H2	H2	0	0	0	1	0	1	EA
H1	H1	0	1	0	0	-1	1	EA
EA	EA	0	0	1	0	0	0	EA
YC	YC	0	0	0	0	0	0	EA
Y2	Y2	0	0	0	0	0	0	EA
CC	CC	0	1	0	0	0	0	EA
CA	CA	0	-1	1	1	0	0	EA
H4	H4	0	0	0	0	0	0	EA
CE	CE	0	0	-1	0	0	0	EA
EB	EB	0	0	0	0	0	0	EA
H5	H5	0	0	0	0	-1	0	EA
CD	CD	0	0	0	0	0	0	EA

```
BOX PLOT OF  180 STANDERSIZED RESIDUALS FROM LPLT= -2.237    TO ULPT=  1.961
NO.<LPLT                                                          NO.>UPLT
       1 *  **  *------------------I     +     I-------------     ***   0
```

ANALYSIS OF VARIANCE FOR THE AMMI MODEL

SOURCE	D.F.	S.S.	M.S.	F	FPROB
TREATMENTS	19	0.136225E+08	716976.		
LOCATIONS	8	0.415143E+08	0.518928E+07		
TREATMENT X SITES	152	0.196454E+08	129246.		
AMMI COMPONENT 1	26	0.102631E+08	394736.	5.301	0.000
AMMI COMPONENT 2	24	0.310145E+07	129227.	2.099	0.006
AMMI COMPONENT 3	22	0.251170E+07	114168.	2.423	0.002
AMMI COMPONENT 4	20	0.127500E+07	63750.1	1.534	0.103
GXE RESIDUAL	60	0.249409E+07			
TOTAL	179	0.747822E+08			

 GENOTYPE MAP SHOWING BEST GENOTYPES OVER THE RANGE OF AMMI-2 SITE SCORES (2 CHRS/PIXEL)

```
    33.74 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    32.80 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    31.86 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    30.92 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    29.99 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    29.05 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    28.11 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    27.17 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    26.24 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    25.30 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    24.36 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    23.42 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    22.49 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    21.55 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    20.61 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    19.68 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    18.74 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    17.80 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    16.86 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    15.93 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    14.99 CDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    14.05 CDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    13.11 CDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    12.18 CDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    11.24 CDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6
    10.30 CDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6
     9.36 CDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6
     8.43 CDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6
     7.49 CDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6H6
     6.55 CDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6H6H6
     5.61 CDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6H6H6
     4.68 CDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6H6H6H6
     3.74 CDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6H6H6H6H6
     2.80 CDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6H6H6H6H6
     1.86 CDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6H6H6H6H6H6H6
     0.93 CDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6H6H6H6H6H6H6H6
```

```
 -0.01 CDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6H6H6H6H6H6H6H6H6
 -0.95 CDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6H6H6H6H6H6H6H6H6
 -1.89 CDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6H6H6H6H6H6H6H6H6
 -2.82 CDCDCDCDH3H3G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6H6H6H6H6H6H6H6H6
 -3.76 CDCDCDCDH3H3H3H3H3H3H3H3H3H3G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H6H6H6H6H6H6H6H6H6H6H6H6
 -4.70 CDCDCDCDH3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H1H1H1H1H6H6H6H6H6H6H6H6H6H6H6
 -5.64 CDCDCDCDH3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3D2D2D2D2D2D2H1H1H1H1H1H1H1H1H1H1H1H1H1H6H6H6
 -6.57 CDCDCDCDH3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3D2D2D2H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
 -7.51 CDCDCDH3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
 -8.45 CDCDCDH3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
 -9.38 CDCDCDH3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
-10.32 CDCDCDH3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
-11.26 CDCDCDH3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
-12.20 CDCDCDH3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
-13.13 CDCDCDH3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
-14.07 CDCDH3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
-15.01 CDCDH3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
-15.95 CDCDH3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3H3EAEAEAEAEAH1H1H1H1H1H1H1H1H1H1H1H1H1H1
        ^           ^           ^           ^           ^           ^           ^           ^           ^
    -0.258E+02  -0.184E+02  -0.110E+02  -0.359E+01   0.380E+01   0.112E+02   0.186E+02   0.260E+02   0.333E+02
```

AMMI RESIDUALS, ADDITIVE EFFECTS AND MULTIPLICATIVE SCORES FILE 07_ELITE   16/ 7/ 9 0:33
 :PAGE 10

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
CA	CA	87.84	68.09	344.3	144.6	-74.89	124.0	442.8
CC	CC	-206.9	37.27	-79.90	91.35	86.14	-151.8	-73.71
CE	CE	147.0	65.78	-480.7	170.4	30.70	-95.26	-112.4
CD	CD	-35.18	48.39	-8.902	-87.48	-28.94	81.72	14.90
D1	D1	-9.158	-144.6	41.35	259.5	49.74	114.2	18.04
D2	D2	-227.7	128.7	-136.9	140.2	153.8	-278.5	-97.23
D3	D3	-18.92	14.38	-89.17	106.3	54.22	11.46	-38.62
D4	D4	-345.2	331.8	-106.0	-32.48	89.28	-77.27	52.81
YC	YC	10.05	123.8	-163.7	-290.1	-79.74	219.9	-72.17
Y2	Y2	96.00	-51.21	134.0	85.08	-88.50	143.1	221.5
EA	EA	-195.0	-191.4	260.5	175.6	116.5	53.60	-70.21
EB	EB	-225.6	100.5	25.33	45.60	103.0	-83.69	-29.86
G1	G1	-49.75	-207.7	327.0	-79.34	37.53	-95.89	-115.0
G2	G2	154.4	-480.7	118.7	61.49	0.2047	110.1	-277.8
H1	H1	257.9	-149.0	224.7	-135.3	-131.1	-248.2	94.06
H2	H2	258.1	261.3	-132.2	-245.2	-189.9	-170.5	250.4
H3	H3	-88.28	-116.9	0.3079	9.993	32.99	338.3	-134.6
H4	H4	71.69	207.8	-94.87	-498.3 *	-99.07	137.6	-92.81
H5	H5	212.4	-142.9	-71.62	194.7	24.34	-424.8	-46.48
H6	H6	106.5	96.74	-112.4	-116.6	-86.18	291.8	66.35
SITE EFFECTS		-77.69	-405.1	261.3 ***	-1003. ***	803.8 ***	68.54 ***	128.2 ***
AMMI1 SITE		-19.71	-25.75 ***	-4.717	-.5641E-01	-15.16	5.013 ***	39.50 ***
AMMI2 SITE		3.899	-10.93 ***	-10.21	-15.95 ***	33.74 ***	-2.422	9.444

SECTION 2

VARIETY\SITE		RI 2007		AL 2007		T-EFCTS		AMMI1 TRT		AMMI2 TRT	
CA	CA	-731.2	**	-405.6		-70.20	***	-4.764		9.201	***
CC	CC	32.00		265.5		-10.97		8.008	***	1.879	
CE	CE	486.7	*	-212.1		-120.4	***	0.2399		-7.075	***
CD	CD	-71.04		86.55		-853.5		-50.16		0.7336	
D1	D1	-221.4		-107.6		168.3		-.4452	***	-3.989	***
D2	D2	334.6		-16.98		235.6	***	3.832	***	-.1953	
D3	D3	145.0		-184.6		113.5	***	8.684		14.10	
D4	D4	42.22		44.89		215.9	***	4.254		12.05	
YC	YC	162.3		89.74		74.53		-.3021		7.933	***
Y2	Y2	-555.1	*	15.09		18.94	***	8.344		-11.21	***
EA	EA	-197.5		47.95		84.91	***	2.234	***	-16.39	***
EB	EB	86.05		-21.25		-389.9	***	0.6095	***	-10.61	***
G1	G1	11.51		171.6		219.5	***	1.939	***	11.18	
G2	G2	39.87		273.7		317.2	***	3.469	***	14.42	
H1	H1	-177.6		264.6		100.5		8.901		-8.902	
H2	H2	-44.99		12.97		101.1		6.292	***	0.5847	***
H3	H3	-34.71		-7.123		206.8	***	1.230	***	-9.353	
H4	H4	466.4		-98.39		-93.75		0.8837		-12.04	***
H5	H5	261.2		-6.791		-463.8		-13.36		0.8579	
H6	H6	-34.13		-212.1		145.9	***	10.11		6.826	
SITE EFFECTS		-151.7	***	375.7		3047.	***	.		.	
AMMI1 SITE		17.38	***	3.505		.		.		.	
AMMI2 SITE		-5.516	***	-2.054		.		.		.	

FITTED VALUES FROM THE AMMI MODEL FILE 07_ELITE   16/ 7/ 9 0:33
---------- :PAGE 11

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
CA	CA	3029.	2594.	3167.	1828.	4163.	2999.	3004.
CC	CC	2808.	2404.	3241.	2003.	3782.	3140.	3499.
CE	CE	2817.	2593.	3259.	2037.	3488.	3014.	2998.
CD	CD	3108.	3072.	2684.	1182.	3783.	2009.	347.8
D1	D1	3131.	2866.	3520.	2276.	3892.	3292.	3289.
D2	D2	3129.	2781.	3528.	2283.	4022.	3371.	3561.
D3	D3	2967.	2378.	3237.	1932.	4309.	3239.	3765.
D4	D4	3149.	2617.	3381.	2068.	4409.	3324.	3673.
YC	YC	3081.	2638.	3304.	1992.	4198.	3170.	3313.
Y2	Y2	2780.	2569.	3403.	2242.	3365.	3204.	3418.
EA	EA	2947.	2849.	3550.	2390.	3349.	3252.	3194.
EB	EB	2526.	2353.	3024.	1824.	3094.	2755.	2709.
G1	G1	3194.	2690.	3405.	2085.	4418.	3318.	3577.
G2	G2	3275.	2712.	3462.	2131.	4602.	3415.	3766.
H1	H1	2860.	2611.	3458.	2286.	3516.	3283.	3544.
H2	H2	2949.	2575.	3374.	2136.	3876.	3247.	3531.
H3	H3	3116.	2920.	3605.	2400.	3724.	3351.	3343.
H4	H4	2811.	2657.	3334.	2143.	3338.	3056.	3003.
H5	H5	2773.	2513.	2899.	1568.	3619.	2583.	2192.
H6	H6	2943.	2453.	3337.	2081.	4074.	3296.	3785.
SITE ESTS.		2970.	2642.	3309.	2044.	3851.	3116.	3176.
AMMI1 SITE		-19.71	-25.75	-4.717	-.5641E-01	-15.16	5.013	39.50
AMMI2 SITE		3.899 I	-10.93 I	-10.21 I	-15.95 I	33.74 I	-2.422 I	9.444 I

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	T-ESTS.	AMMI1 TRT	AMMI2 TRT
CA	CA	2692.	3317.	2977.	-4.764	9.201 I
CC	CC	3014.	3436.	3036.	8.008	1.879 I
CE	CE	2818.	3318.	2927.	0.2399	-7.075 I
CD	CD	1166.	2392.	2194.	-50.16	0.7336 I
D1	D1	3078.	3598.	3216.	-.4452	-3.989 I
D2	D2	3199.	3672.	3283.	3.832	-.1953 I
D3	D3	3082.	3538.	3161.	8.684	14.10 I
D4	D4	3119.	3629.	3263.	4.254	12.05 I
YC	YC	2921.	3480.	3122.	-.3021	7.933 I
Y2	Y2	3121.	3494.	3066.	8.344	-11.21 I
EA	EA	3110.	3549.	3132.	2.234	-16.39 I
EB	EB	2575.	3057.	2657.	0.6095	-10.61 I
G1	G1	3087.	3626.	3267.	1.939	11.18 I
G2	G2	3194.	3723.	3364.	3.469	14.42 I
H1	H1	3200.	3573.	3148.	8.901	-8.902 I
H2	H2	3103.	3545.	3148.	6.292	0.5847 I
H3	H3	3175.	3653.	3254.	1.230	-9.353 I
H4	H4	2884.	3357.	2954.	0.8837	-12.04 I
H5	H5	2195.	2911.	2583.	-13.36	0.8579 I
H6	H6	3180.	3590.	3193.	10.11	6.826 I
SITE ESTS.		2896.	3423.	3047.	.	.
AMMI1 SITE		17.38	3.505	.	.	.
AMMI2 SITE		-5.516 I	-2.054 I	.	.	.

Addendum 8: Cross-site analysis of total starch yield, combined 2006-2007 seasons data

```
PBGXE - CROSS SITE ANALYSIS  FILE 0607_STY   23/ 7/ 9 15: 6
 ------------------------------------------------------------------ :PAGE   1

   7       CN$    CODES:
  1 CA          CA            2 CC          CC            3 CD          CD
  4 CE          CE            5 D1          D1            6 YC          YC
  7 Y2          Y2

  15     SITE$    CODES:
  1 VR 2006       2 LA 2006       3 ME 2006       4 RO 2006       5 TY 2006
  6 NA 2006       7 PI 2007       8 KL 2007       9 LA 2007      10 ME 2007
 11 RO 2007      12 TY 2007      13 NA 2007      14 RI 2007      15 AL 2007

 TREATMENT BY VARIATE MEANS HAVE BEEN REQUESTED FOR  1 VARIATES:
 STY_KGHA
 ROWS OF MEANS TABLES TO BE SORTED ON VARIATE STY_KGHA
 GXE ANALYSIS HAS BEEN REQUESTED FOR  1 VARIATES


TREATMENT MEANS AND COUNTS OVER SITES FOR EACH VARIATE.  FILE 0607_STY   23/ 7/ 9 15: 6
 ------------------------------------------------------------------------------ :PAGE   2
 VARIETY\SITE             |STY_KGHA    |
 -------------------------|------------
 YC          YC           | 2752.    51|
 CA          CA           | 2703.    51|
 D1          D1           | 2702.    51|
 Y2          Y2           | 2687.    51|
 CC          CC           | 2685.    51|
                          |            |
 CE          CE           | 2573.    51|
 CD          CD           | 2144.    51|
 -------------------------|------------
 SITE MEANS               | 2607.   357|
```

7 X 15 MATRIX OF TREATMENT BY SITE MEANS FOR VARIATE STY_KGHA FILE 0607_STY 23/ 7/ 9 15: 6
:PAGE 3

SECTION 1

VARIETY\SITE		VR 2006		LA 2006		ME 2006		RO 2006		TY 2006		NA 2006		PI 2007	
YC	YC	2098.	4	1536.	4	1407.	4	3489.	4	2633.	4	2852.	4	3091.	3
CA	CA	1786.	4	2017.	4	1638.	4	3577.	4	2251.	4	3104.	4	3117.	3
D1	D1	1670.	4	1384.	4	1706.	4	3147.	4	1952.	4	2886.	4	3122.	3
Y2	Y2	1584.	4	1735.	4	1494.	4	3001.	4	2667.	4	3081.	4	2876.	3
CC	CC	1880.	4	1599.	4	1313.	4	3228.	4	2301.	4	3412.	4	2601.	3
CE	CE	1665.	4	1604.	4	1696.	4	3258.	4	2124.	4	2695.	4	2964.	3
CD	CD	2186.	4	2007.	4	1161.	4	3213.	4	1566.	4	2399.	4	3073.	3
SITE MEANS		1839.	28	1697.	28	1488.	28	3273.	28	2214.	28	2919.	28	2978.	21
SITE INDEX		1839.	28	1697.	28	1488.	28	3273.	28	2214.	28	2919.	28	2978.	21

SECTION 2

VARIETY\SITE		KL 2007		LA 2007		ME 2007		RO 2007		TY 2007		NA 2007		RI 2007	
YC	YC	2762.	3	3140.	3	1702.	3	4118.	3	3390.	3	3241.	3	3083.	3
CA	CA	2662.	3	3511.	3	1972.	3	4089.	3	3123.	3	3447.	3	1961.	3
D1	D1	2721.	3	3561.	3	2536.	3	3941.	3	3406.	3	3307.	3	2857.	3
Y2	Y2	2518.	3	3537.	3	2327.	3	3277.	3	3347.	3	3640.	3	2566.	3
CC	CC	2442.	3	3161.	3	2094.	3	3868.	3	2989.	3	3425.	3	3046.	3
CE	CE	2659.	3	2779.	3	2207.	3	3519.	3	2919.	3	2885.	3	3305.	3
CD	CD	3121.	3	2675.	3	1094.	3	3754.	3	2091.	3	362.7	3	1095.	3
SITE MEANS		2698.	21	3195.	21	1990.	21	3795.	21	3038.	21	2901.	21	2559.	21
SITE INDEX		2698.	21	3195.	21	1990.	21	3795.	21	3038.	21	2901.	21	2559.	21

SECTION 3

VARIETY\SITE		AL 2007		TRT MEANS	
YC	YC	3570.	3	2752.	51
CA	CA	2912.	3	2703.	51
D1	D1	3490.	3	2702.	51
Y2	Y2	3509.	3	2687.	51
CC	CC	3702.	3	2685.	51
CE	CE	3106.	3	2573.	51
CD	CD	2479.	3	2144.	51
SITE MEANS		3253.	21	2607.	357
SITE INDEX		3253.	21	2607.	357

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

ENVIRONMENT	MEAN	SE	DUNCAN GROUPS	LSD TESTS
RO 2007	3795.1	165.82	|	.
RO 2006	3273.2	143.60	||	1.
AL 2007	3252.5	165.82	|||	1..
LA 2007	3194.7	165.82	||	1...
TY 2007	3037.7	165.82	|||	2....
PI 2007	2977.7	165.82	|||	3.....
NA 2006	2918.5	143.60	|||	3.....
NA 2007	2901.0	165.82	|||	3.....
KL 2007	2697.6	165.82	|||	3111.....
RI 2007	2559.0	165.82	|||	32221.....
TY 2006	2213.5	143.60	|||	333333321..
ME 2007	1990.4	165.82	|||	3333333321..
VR 2006	1838.7	143.60	||	3333333332...
LA 2006	1697.4	143.60	|	33333333331...
ME 2006	1487.8	143.60	|	333333333331...

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

TREATMENT		MEAN	SE	DUNCAN GROUPS	LSD TESTS
YC	YC	2801.1	106.56	|	.
CA	CA	2752.5	106.56	|	..
D1	D1	2751.0	106.56	|	...
Y2	Y2	2736.2	106.56	|	....
CC	CC	2733.7	106.56	|	.....
CE	CE	2621.6	106.56	|	.....
CD	CD	2193.5	106.56		333332.

RESIDUALS FROM THE ADDITIVE TREATMENT BY SITE MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN STANDARD ERRORS,
 ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		R O 2 0 0 7	R O 2 0 0 6	A L 2 0 0 7	L A 2 0 0 7	T Y 2 0 0 7	P I 2 0 0 7	N A 2 0 0 6	N A 2 0 0 7	K L 2 0 0 7	R I 2 0 0 7	T Y 2 0 0 6	M E 2 0 0 7	V R 2 0 0 6	L A 2 0 0 6	M E 2 0 0 6	T - E F C T S
YC	YC	0	0	0	0	0	0	0	0	0	0	0	-1	0	0	0	1
CA	CA	0	0	-1	0	0	0	0	1	0	-1	0	0	0	0	0	0
D1	D1	0	0	0	0	0	0	0	0	0	0	-1	1	0	-1	0	0
Y2	Y2	-1	-1	0	0	0	0	0	1	0	0	1	0	0	0	0	0
CC	CC	0	0	0	0	0	-1	1	1	0	1	0	0	0	0	0	0
CE	CE	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0
CD	CD	1	1	0	0	-1	1	0	-5	2	-2	0	-1	2	2	0	-4
SITE EFFECTS		7	4	0	3	2	2	1	1	0	0	-3	-4	-5	-6	-8	65

BOX PLOT OF 105 STUDENTIZED RESIDUALS FROM LPLT= -2.542   TO ULPT= 2.398

```
NO.<LPLT                                                        NO.>UPLT
      1 *        --------------I     +      I-----------------  ***   0
```

MEDIAN= -0.1253E-01 ANDERSON-DARLING STATISTIC= 1.088 **

ANALYSIS OF VARIANCE FOR THE ADDITIVE MODEL

SOURCE	D.F.	S.S.	M.S.	F	FPROB
TREATMENTS	6	0.400441E+07	667402.		
LOCATIONS	14	0.460860E+08	0.329186E+07		
TREATMENT X SITES	84	0.142657E+08	169830.		
TOTAL	104	0.643562E+08			

RESIDUALS FROM THE ADDITIVE SITE X TREATMENT MODEL FILE 0607_STY   23/ 7/ 9 15: 6
 -- :PAGE   5

 SECTION 1

VARIETY\SITE		VR 2006	LA 2006	ME 2006	RO 2006	TY 2006	NA 2006	PI 2007
YC	YC	123.8	-332.7	-245.4	75.89	297.4	-229.4	-30.15
CA	CA	-161.8	241.4	57.64	224.2	-64.41	96.18	39.90
D1	D1	-286.4	-443.6	132.8	-240.7	-386.9	-139.0	45.93
Y2	Y2	-363.0	-46.46	-80.69	-382.5	405.0	88.47	-170.8
CC	CC	-39.67	-191.8	-274.1	-133.7	10.14	451.1	-427.0
CE	CE	-151.2	-63.95	263.2	20.59	-60.31	-205.6	18.93
CD	CD	878.3 *	837.1 *	146.5	436.3	-200.9	-61.67	523.2
SITE EFFECTS		-816.9 ***	-958.2 ***	-1168. ***	617.5 ***	-442.2 **	262.8	322.1 *

 SECTION 2

VARIETY\SITE		KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007	RI 2007
YC	YC	-76.59	-188.2	-407.3	166.7	193.9	182.6	356.0
CA	CA	-124.3	206.3	-108.0	184.7	-10.37	421.8	-653.0
D1	D1	-67.67	254.5	422.7	47.72	256.2	291.4	190.1
Y2	Y2	-244.7	245.5	240.3	-562.5	214.9	618.2	-68.74
CC	CC	-313.7	-105.3	24.29	-4.639	-119.3	418.8	383.7
CE	CE	-4.558	-359.0	235.5	-227.4	-79.98	17.27	732.8
CD	CD	831.4 *	-53.89	-407.4	395.3	-455.3	-1950. ***	-940.9 *
SITE EFFECTS		41.94	539.1 **	-665.3 ***	1139. ***	382.0 *	245.3	-96.65

SECTION 3

VARIETY\SITE		AL 2007	T-EFCTS	
YC	YC	161.5	145.5	
CA	CA	-411.2	96.85	
D1	D1	133.8	95.34	
Y2	Y2	165.6	80.49	
CC	CC	348.7	78.06	
CE	CE	-105.8	-34.03	
CD	CD	-292.7	-462.2	***
SITE EFFECTS		596.8	2656.	***

FITTED VALUES FROM ADDITIVE SITE X TREATMENT MODEL FILE 0607_STY   23/ 7/ 9 15: 6
 -- :PAGE   6

 SECTION 1

VARIETY\SITE		VR 2006	LA 2006	ME 2006	RO 2006	TY 2006	NA 2006	PI 2007
YC	YC	1984.	1843.	1633.	3419.	2359.	3064.	3123.
CA	CA	1936.	1794.	1585.	3370.	2310.	3015.	3075.
D1	D1	1934.	1793.	1583.	3369.	2309.	3014.	3073.
Y2	Y2	1919.	1778.	1568.	3354.	2294.	2999.	3058.
CC	CC	1917.	1775.	1566.	3351.	2292.	2997.	3056.
CE	CE	1805.	1663.	1454.	3239.	2179.	2884.	2944.
CD	CD	1377.	1235.	1026.	2811.	1751.	2456.	2516.
SITE ESTS.		1839.	1697.	1488.	3273.	2214.	2919.	2978.

 SECTION 2

VARIETY\SITE		KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007	RI 2007
YC	YC	2843.	3340.	2136.	3941.	3183.	3046.	2704.
CA	CA	2794.	3292.	2087.	3892.	3135.	2998.	2656.
D1	D1	2793.	3290.	2086.	3890.	3133.	2996.	2654.
Y2	Y2	2778.	3275.	2071.	3876.	3118.	2981.	2640.
CC	CC	2776.	3273.	2068.	3873.	3116.	2979.	2637.
CE	CE	2664.	3161.	1956.	3761.	3004.	2867.	2525.
CD	CD	2235.	2733.	1528.	3333.	2576.	2439.	2097.
SITE ESTS.		2698.	3195.	1990.	3795.	3038.	2901.	2559.

SECTION 3

VARIETY\SITE		AL 2007	T-ESTS.
YC	YC	3398.	2801.
CA	CA	3349.	2753.
D1	D1	3348.	2751.
Y2	Y2	3333.	2736.
CC	CC	3331.	2734.
CE	CE	3218.	2622.
CD	CD	2790.	2193.
SITE ESTS.		3253.	2656.

 REGRESSIONS OF STY_KGHA FOR EACH VARIETY ON MEANS OF STY_KGHA AT EACH SITE

VARIETY		MEAN	SLOPE	SE	MS-TXL	MS-REG	MS-DEV	R**2(%)
CA	CA	2752.51	1.029	0.112	73595.98	5293.10	78850.04	1.
CC	CC	2733.72	1.102	0.112	77556.17	65013.36	78520.99	6.
CD	CD	2193.49	0.679	0.299	571416.00	650591.38	565325.56	8.
CE	CE	2621.63	0.870	0.100	66707.14	106630.40	63636.12	11.
D1	D1	2751.00	1.191	0.096	70032.30	229658.28	57753.37	23.
YC	YC	2801.12	1.161	0.088	56625.26	163201.67	48427.08	21.
Y2	Y2	2736.15	0.969	0.132	103047.46	6054.15	110508.49	0.

 SLOPE - SLOPES OF REGRESSIONS OF VARIETY MEANS ON SITE INDEX.
          * INDICATES SLOPES SIGNIFICANTLY DIFFERENT FROM THE
          SLOPE FOR THE OVERALL REGRESSION WHICH IS   1.00
 MS-TXL - CONTRIBUTION OF EACH VARIETY TO INTERACTION MS
 MS-REG - CONTRIBUTION OF EACH VARIETY TO THE REGRESSION
          COMPONENT OF THE TREATMENT BY LOCATION INTERACTION
 MS-DEV - DEVIATIONS FROM REGRESSION COMPONENT OF INTERACTION
 R**2   - SQUARED CORRELATION BETWEEN RESIDUALS FROM THE MAIN
          EFFECTS MODEL AND THE SITE INDEX.
 VARIATE STY_KGHA WAS SITE INDEX WITH OVERALL MEAN 2607.
 THE FOLLOWING SITE MEANS OF STY_KGHA WERE USED AS X-VARIATES
           1839.   1697.   1488.   3273.   2214.   2919.   2978.   2698.   3195.   1990.
           3795.   3038.   2901.   2559.   3253.

 ANOVA FOR VARIABLE STY_KGHA WITH SITE REGRESSIONS ON STY_KGHA

SOURCE	D.F.	S.S.	M.S.	F	FPROB
TREATMENTS	6	0.400441E+07	667402.		
LOCATIONS	14	0.460860E+08	0.329186E+07		
TREATMENT X SITES	84	0.142657E+08	169830.		
TRT X SITE REG	6	0.122644E+07	204407.	1.223	0.303
DEVIATIONS	78	0.130393E+08	167170.		
TOTAL	104	0.643562E+08			

```
AMMI ANALYSIS  FILE 0607_STY   23/ 7/ 9 15: 6
 ------------------------------------------------------------- :PAGE   8
 SINGULAR VALUES OF INTERACTION MATRIX (CONDITION=  0)
 3153.3    1309.1    1004.5    798.43    713.55    672.97

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR TREATMENTS

 CA CA          CA           0.152643E+01 0.248725E+02-0.751691E+00 0.127771E+02
 CC CC          CC          -0.131706E+02-0.490736E+01-0.147567E+02-0.692360E+01
 CD CD          CD           0.502579E+02-0.148705E+01 0.159190E+00-0.549326E+01
 CE CE          CE          -0.590915E+01-0.190361E+02 0.120778E+02-0.165616E+01
 D1 D1          D1          -0.109468E+02-0.971039E+00 0.192251E+02 0.739934E+01
 YC YC          YC          -0.539099E+01-0.114756E+02-0.164448E+02 0.124554E+02
 Y2 Y2          Y2          -0.163668E+02 0.130047E+02 0.491067E+00-0.185588E+02

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR ENVIRONMENTS

 A              VR 2006      0.170359E+02-0.620475E+01-0.865967E+01-0.259405E+00
 B              LA 2006      0.167293E+02 0.806915E+01-0.106502E+01-0.832208E+01
 C              ME 2006      0.339125E+01-0.619960E+00 0.136906E+02 0.102400E+01
 D              RO 2006      0.102733E+02-0.320401E+00-0.392351E+01 0.954771E+01
 E              TY 2006     -0.443079E+01 0.154719E+01-0.129334E+02-0.797237E+01
 F              NA 2006     -0.201950E+01 0.619034E+01-0.804198E+01-0.844520E+01
 G              PI 2007      0.108853E+02 0.224633E-01 0.784312E+01 0.462822E+01
 H              KL 2007      0.161454E+02-0.377254E+01 0.461700E+01-0.111473E+01
 I              LA 2007     -0.148277E+01 0.134972E+02 0.513904E+01-0.955204E+00
 J              ME 2007     -0.910711E+01 0.538388E+00 0.173666E+02-0.764608E+01
 K              RO 2007      0.930446E+01-0.700350E+00-0.483235E+01 0.168652E+02
 L              TY 2007     -0.894943E+01 0.217509E+01 0.256105E+01 0.457227E+01
 M              NA 2007     -0.371919E+02 0.127338E+02-0.367918E+01 0.767939E+01
 N              RI 2007     -0.192000E+02-0.273773E+02 0.129178E+01 0.904098E-01
 O              AL 2007     -0.772305E+01-0.711823E+01-0.613573E+01-0.746025E+01
```

RESIDUALS FROM THE AMMI-2 MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN UNITS OF ROOT (RESIDUAL GXE MS), ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		R O 2 0 7	R O 2 0 6	A L 2 0 7	L A 2 0 7	T Y 2 0 7	P I 2 0 7	N A 2 0 6	N A 2 0 7	K L 2 0 7	R I 2 0 7	T Y 2 0 6	M E 2 0 7	V E 2 0 6	L A 2 0 6	M E 2 0 6	T - E S
YC	YC	0	0	0	0	0	0	0	0	0	0	1	-1	0	0	-1	65
CA	CA	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	65
D1	D1	0	0	0	1	0	0	0	0	0	0	-1	1	0	-1	0	65
Y2	Y2	-1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	65
CC	CC	0	0	0	0	0	-1	1	0	0	0	0	0	0	0	0	65
CE	CE	0	0	-1	0	0	0	0	0	0	0	0	0	0	0	1	65
CD	CD	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	65

BOX PLOT OF 105 STANDERSIZED RESIDUALS FROM LPLT= -1.931 TO ULPT= 1.951

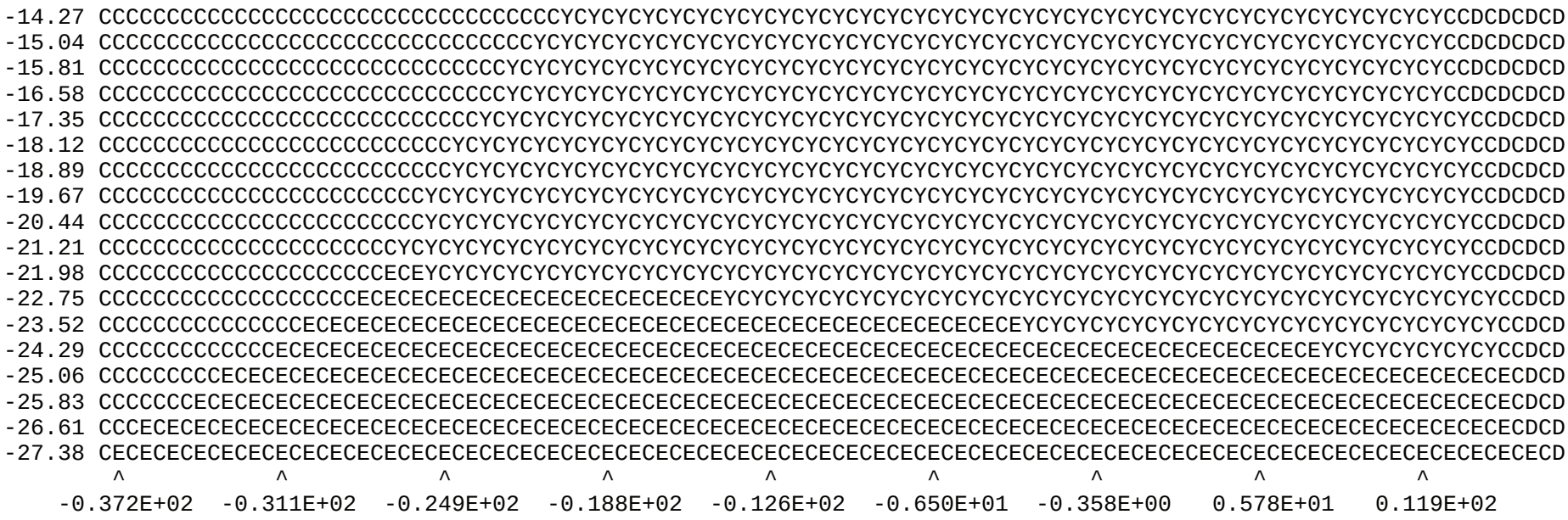
```
NO.<LPLT                                                    NO.>UPLT
       0 ** *       ------------I     +      I-------------        *   0
```

ANALYSIS OF VARIANCE FOR THE AMMI MODEL

SOURCE	D.F.	S.S.	M.S.	F	FPROB
TREATMENTS	6	0.400441E+07	667402.		
LOCATIONS	14	0.460860E+08	0.329186E+07		
TREATMENT X SITES	84	0.142657E+08	169830.		
AMMI COMPONENT 1	19	0.994352E+07	523343.	7.870	0.000
AMMI COMPONENT 2	17	0.171365E+07	100803.	1.855	0.048
AMMI COMPONENT 3	15	0.100902E+07	67268.1	1.388	0.210
AMMI COMPONENT 4	13	637487.	49037.5	1.019	0.471
GXE RESIDUAL	20	962048.			
TOTAL	104	0.643562E+08			

 GENOTYPE MAP SHOWING BEST GENOTYPES OVER THE RANGE OF AMMI-2 SITE SCORES (2 CHRS/PIXEL)

```
 13.50 Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2CACACACACACACACACACACACACACACACACACACACACACACACACACACACACACACA
 12.73 Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2CACACACACACACACACACACACACACACACACACACACACACACACACACACACACACACA
 11.95 Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2CACACACACACACACACACACACACACACACACACACACACACACACACACACACACACACA
 11.18 Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2CACACACACACACACACACACACACACACACACACACACACACACACACACACACACACACA
 10.41 Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2CACACACACACACACACACACACACACACACACACACACACACACACACACACACACACACA
  9.64 Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2CACACACACACACACACACACACACACACACACACACACACACACACACACACACACACACD
  8.87 Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2CACACACACACACACACACACACACACACACACACACACACACACACACACACACACACACD
  8.10 Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2CACACACACACACACACACACACACACACACACACACACACACACACACACACACACACDCD
  7.33 Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2CACACACACACACACACACACACACACACACACACACACACACACACACACACACACACDCD
  6.56 Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2CACACACACACACACACACACACACACACACACACACACACACACACACACACACACACDCD
  5.79 Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2CACACACACACACACACACACACACACACACACACACACACACACACACACACACACACDCDCD
  5.01 Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2CACACACACACACACACACACACACACACACACACACACACACACACACACACACACACDCDCD
  4.24 Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2Y2
```


-14.27 CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCCDCDCDCD
-15.04 CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCCDCDCDCD
-15.81 CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCCDCDCDCD
-16.58 CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCCDCDCDCD
-17.35 CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCCDCDCDCD
-18.12 CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCCDCDCDCD
-18.89 CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCCDCDCDCD
-19.67 CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCCDCDCDCD
-20.44 CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCCDCDCDCD
-21.21 CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCCDCDCDCD
-21.98 CCCCCCCCCCCCCCCCCCCCCCCCCECEYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCCDCDCDCD
-22.75 CCCCCCCCCCCCCCCCCCCCCECECECECECECECECECECECECECEYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCYCCDCD
-23.52 CCCCCCCCCCCCCCCCECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECEYCYCYCYCYCYCYCYCYCYCYCYCYCYCCDCD
-24.29 CCCCCCCCCCCCCECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECEYCYCYCYCYCYCCDCD
-25.06 CCCCCCCCECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECDCD
-25.83 CCCCCCECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECDCD
-26.61 CCCECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECDCD
-27.38 CECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECECD
^ ^ ^ ^ ^ ^ ^ ^ ^
-0.372E+02 -0.311E+02 -0.249E+02 -0.188E+02 -0.126E+02 -0.650E+01 -0.358E+00 0.578E+01 0.119E+02

SECTION 1

VARIETY\SITE		VR 2006	LA 2006	ME 2006	RO 2006	TY 2006	NA 2006	PI 2007
YC	YC	144.4	-149.9	-234.2	127.6	291.3	-169.3	28.79
CA	CA	-33.50	15.18	67.88	216.5	-96.13	-54.70	22.73
D1	D1	-105.9	-252.6	169.4	-128.6	-433.9	-155.1	165.1
Y2	Y2	-3.528	122.4	-17.12	-210.2	312.4	-25.08	7.046
CC	CC	154.3	68.16	-232.5	0.1092E-01	-40.62	454.9	-283.5
CE	CE	-168.6	188.5	271.4	75.20	-57.04	-99.74	83.69
CD	CD	12.89	8.318	-24.87	-80.51	24.04	49.03	-23.81
SITE EFFECTS		-816.9	-958.2	-1168.	617.5	-442.2 ***	262.8	322.1
AMMI1 SITE		17.04	16.73	3.391 ***	10.27 ***	-4.431	-2.020 ***	10.89
AMMI2 SITE		-6.205	8.069	-.6200	-.3204	1.547 ***	6.190 ***	0.2246E-01

SECTION 2

VARIETY\SITE		KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007	RI 2007
YC	YC	-32.85	-41.33	-450.2	208.8	170.6	128.2	-61.68
CA	CA	-55.09	-127.1	-107.5	187.9	-50.81	161.9	57.28
D1	D1	105.4	251.4	323.5	148.9	160.4	-103.3	-46.63
Y2	Y2	68.65	45.75	84.21	-401.1	40.11	-156.1	-26.95
CC	CC	-119.5	-58.55	-93.01	114.5	-226.5	-8.557	-3.555
CE	CE	19.03	-110.8	192.0	-185.7	-91.46	39.90	98.21
CD	CD	14.38	40.70	51.06	-73.33	-2.284	-62.05	-16.67
SITE EFFECTS		41.94	539.1 ***	-665.3 ***	1139.	382.0	245.3	-96.65
AMMI1 SITE		16.15 ***	-1.483 ***	-9.107 ***	9.304	-8.949 ***	-37.19	-19.20
AMMI2 SITE		-3.773	13.50 ***	0.5384	-.7003 ***	2.175	12.73 ***	-27.38

SECTION 3

VARIETY\SITE		AL 2007		T-EFCTS		AMMI1 TRT		AMMI2 TRT	
YC	YC	38.21		145.5		-5.391	***	-11.48	***
CA	CA	-222.3		96.85		1.526		24.87	***
D1	D1	42.38		95.34	***	-10.95		-.9710	***
Y2	Y2	131.8		80.49	***	-16.37		13.00	***
CC	CC	212.1		78.06	***	-13.17		-4.907	
CE	CE	-287.0		-34.03		-5.909	***	-19.04	***
CD	CD	84.84		-462.2	***	50.26	***	-1.487	***
SITE EFFECTS		596.8	***	2656.		.		.	
AMMI1 SITE		-7.723		.		.		.	
AMMI2 SITE		-7.118		.		.		.	

FITTED VALUES FROM THE AMMI MODEL FILE 0607_STY   23/ 7/ 9 15: 6
---------- :PAGE 11

SECTION 1

VARIETY\SITE		VR 2006	LA 2006	ME 2006	RO 2006	TY 2006	NA 2006	PI 2007
YC	YC	1964.	1660.	1622.	3367.	2365.	3004.	3064.
CA	CA	1807.	2021.	1574.	3378.	2342.	3166.	3092.
D1	D1	1754.	1602.	1547.	3256.	2356.	3030.	2954.
Y2	Y2	1560.	1609.	1505.	3181.	2387.	3113.	2880.
CC	CC	1723.	1516.	1524.	3218.	2342.	2993.	2912.
CE	CE	1822.	1411.	1446.	3185.	2176.	2779.	2879.
CD	CD	2242.	2064.	1197.	3328.	1526.	2346.	3063.
SITE ESTS.		1839.	1697.	1488.	3273.	2214.	2919.	2978.
AMMI1 SITE		17.04	16.73	3.391	10.27	-4.431	-2.020	10.89
AMMI2 SITE		-6.205	8.069	-.6200	-.3204	1.547	6.190	0.2246E-01

SECTION 2

VARIETY\SITE		KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007	RI 2007
YC	YC	2799.	3193.	2179.	3898.	3206.	3101.	3122.
CA	CA	2725.	3625.	2087.	3889.	3175.	3258.	1946.
D1	D1	2620.	3293.	2185.	3789.	3229.	3391.	2891.
Y2	Y2	2465.	3475.	2227.	3714.	3293.	3756.	2598.
CC	CC	2582.	3226.	2186.	3754.	3223.	3406.	3024.
CE	CE	2640.	2913.	2000.	3719.	3015.	2844.	3160.
CD	CD	3052.	2638.	1070.	3802.	2122.	550.7	1173.
SITE ESTS.		2698.	3195.	1990.	3795.	3038.	2901.	2559.
AMMI1 SITE		16.15	-1.483	-9.107	9.304	-8.949	-37.19	-19.20
AMMI2 SITE		-3.773	13.50	0.5384	-.7003	2.175	12.73	-27.38

SECTION 3

VARIETY\SITE		AL 2007	T-ESTS.	AMMI1 TRT	AMMI2 TRT
YC	YC	3521.	2801.	-5.391	-11.48 I
CA	CA	3161.	2753.	1.526	24.87 I
D1	D1	3439.	2751.	-10.95	-.9710 I
Y2	Y2	3367.	2736.	-16.37	13.00 I
CC	CC	3467.	2734.	-13.17	-4.907 I
CE	CE	3400.	2622.	-5.909	-19.04 I
CD	CD	2413.	2193.	50.26	-1.487 I
SITE ESTS.		3253.	2656.	.	.
AMMI1 SITE		-7.723	.	.	.
AMMI2 SITE		-7.118 I	.	.	.

Addendum 9: Cross-site analysis of ethanol output, the 2007 season trials

```
PBGXE - CROSS SITE ANALYSIS  FILE 07_ELITE   16/ 8/ 9 12:59
 ------------------------------------------------------------------ :PAGE   1

  20       CN$    CODES:
  1 CA          CA            2 CC          CC            3 CE          CE
  4 CD          CD            5 D1          D1            6 D2          D2
  7 D3          D3            8 D4          D4            9 YC          YC
 10 Y2          Y2           11 EA          EA           12 EB          EB
 13 G1          G1           14 G2          G2           15 H1          H1
 16 H2          H2           17 H3          H3           18 H4          H4
 19 H5          H5           20 H6          H6

   9     SITE$    CODES:
  1 PI 2007       2 KL 2007       3 LA 2007       4 ME 2007       5 RO 2007
  6 TY 2007       7 NA 2007       8 RI 2007       9 AL 2007

 TREATMENT BY VARIATE MEANS HAVE BEEN REQUESTED FOR  1 VARIATES:
 EOLT_ASI
 ROWS OF MEANS TABLES TO BE SORTED ON VARIATE EOLT_ASI
 GXE ANALYSIS HAS BEEN REQUESTED FOR  1 VARIATES
```

TREATMENT MEANS AND COUNTS OVER SITES FOR EACH VARIATE. FILE 07_ELITE 16/ 8/ 9 12:59
:PAGE 2

VARIETY\SITE		EOLT_ASI	
D3	D3	483.0	27
H1	H1	478.7	27
G2	G2	477.3	27
EA	EA	476.8	27
D2	D2	475.9	27
D1	D1	474.8	27
G1	G1	474.8	27
CC	CC	474.2	27
H4	H4	474.2	27
H2	H2	473.3	27
CE	CE	473.2	27
H6	H6	472.1	27
CA	CA	471.8	27
H3	H3	468.7	27
D4	D4	467.7	27
YC	YC	466.5	27
Y2	Y2	466.4	27
H5	H5	464.8	27
CD	CD	462.6	27
EB	EB	455.9	27
SITE MEANS		471.6	540

20 X 9 MATRIX OF TREATMENT BY SITE MEANS FOR VARIATE EOLT_ASI FILE 07_ELITE 16/ 8/ 9 12:59
:PAGE 3

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
D3	D3	516.1	475.9	539.5	445.6	490.1	468.5	437.3
H1	H1	516.5	477.2	551.5	432.1	493.8	494.7	433.0
G2	G2	485.3	531.9	568.7	462.3	494.7	459.4	435.8
EA	EA	510.1	508.9	524.6	444.4	485.9	450.6	457.1
D2	D2	510.8	496.7	509.0	433.7	494.6	503.0	430.6
D1	D1	492.8	526.8	503.1	441.5	501.1	480.7	432.2
G1	G1	470.5	499.5	543.2	442.1	514.3	448.5	435.4
CC	CC	515.2	469.2	528.2	435.2	484.8	486.7	426.9
H4	H4	518.6	458.9	575.2	434.4	464.9	459.3	435.5
H2	H2	480.4	465.9	578.0	443.5	479.2	481.7	437.2
CE	CE	510.5	496.1	507.0	438.7	502.4	456.9	421.9
H6	H6	491.4	469.2	575.1	437.5	483.1	445.4	441.3
CA	CA	485.3	464.9	524.1	434.5	502.9	495.8	425.3
H3	H3	500.8	452.0	495.0	447.9	487.2	491.2	442.3
D4	D4	509.6	476.1	508.3	429.0	501.3	442.9	435.7
YC	YC	488.8	505.8	531.9	430.4	487.7	482.5	415.8
Y2	Y2	487.6	453.0	566.9	447.3	479.6	467.0	432.1
H5	H5	476.4	472.6	553.7	446.2	475.5	447.8	422.9
CD	CD	503.5	514.1	489.8	428.4	429.2	489.6	423.9
EB	EB	451.2	468.6	503.6	443.2	463.3	451.8	424.8
SITE MEANS		496.1	484.2	533.8	439.9	485.8	470.2	432.4
SITE INDEX		496.1	484.2	533.8	439.9	485.8	470.2	432.4

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	TRT MEANS
D3	D3	434.6	539.6	483.0
H1	H1	419.1	490.2	478.7
G2	G2	413.5	444.3	477.3
EA	EA	418.4	490.9	476.8
D2	D2	416.8	487.9	475.9
D1	D1	415.6	479.8	474.8
G1	G1	412.2	507.5	474.8
CC	CC	418.9	502.9	474.2
H4	H4	415.1	505.7	474.2
H2	H2	423.7	470.0	473.3
CE	CE	417.5	507.8	473.2
H6	H6	437.3	468.2	472.1
CA	CA	410.6	502.8	471.8
H3	H3	430.0	471.5	468.7
D4	D4	411.2	494.8	467.7
YC	YC	403.3	452.3	466.5
Y2	Y2	419.3	444.6	466.4
H5	H5	414.6	473.2	464.8
CD	CD	403.2	481.7	462.6
EB	EB	402.6	494.2	455.9
SITE MEANS		416.9	485.5	471.6
SITE INDEX		416.9	485.5	471.6

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

ENVIRONMENT	MEAN	SE	DUNCAN GROUPS	LSD TESTS
LA 2007	533.81	4.2636		.
PI 2007	496.07	4.2636	|	3.
RO 2007	485.78	4.2636	|	3..
AL 2007	485.50	4.2636	|	3...
KL 2007	484.17	4.2636	|	3....
TY 2007	470.19	4.2636		33111.
ME 2007	439.90	4.2636	|	333333.
NA 2007	432.36	4.2636	|	333333..
RI 2007	416.86	4.2636		33333331.

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

TREATMENT		MEAN	SE	DUNCAN GROUPS	LSD TESTS
D3	D3	483.03	6.3557	|	.
H1	H1	478.67	6.3557	|	..
G2	G2	477.31	6.3557	||	...
EA	EA	476.76	6.3557	||	....
D2	D2	475.89	6.3557	||	.....
D1	D1	474.84	6.3557	||	.....
G1	G1	474.79	6.3557	||	.....
CC	CC	474.23	6.3557	||	.....
H4	H4	474.18	6.3557	||	.....
H2	H2	473.30	6.3557	||	.....
CE	CE	473.22	6.3557	||	.....
H6	H6	472.05	6.3557	||	.....
CA	CA	471.78	6.3557	||	.....
H3	H3	468.66	6.3557	||	.....
D4	D4	467.65	6.3557	||	.....
YC	YC	466.50	6.3557	||	.....
Y2	Y2	466.39	6.3557	||	.....
H5	H5	464.77	6.3557	||	1.....
CD	CD	462.60	6.3557	||	1.....
EB	EB	455.93	6.3557	|	211111111.....

RESIDUALS FROM THE ADDITIVE TREATMENT BY SITE MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN STANDARD ERRORS, ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		LA	PIO	RAL	KLY	TEA	MAI	NA	RI	T-EFCTS
		2	2	2	2	2	2	2	2	
		0	0	0	0	0	0	0	0	
		0	0	0	0	0	0	0	0	
		7	7	7	7	7	7	7	7	
D3	D3	0	0	0	2	-1	0	0	0	0	1
H1	H1	0	0	0	0	0	0	0	0	0	1
G2	G2	1	0	0	-2	2	0	0	0	0	0
EA	EA	0	0	0	0	1	-1	0	1	0	0
D2	D2	-1	0	0	0	0	1	0	0	0	0
D1	D1	-1	0	0	0	2	0	0	0	0	0
G1	G1	0	-1	1	1	0	-1	0	0	0	0
CC	CC	0	0	0	0	-1	0	0	0	0	0
H4	H4	2	1	-1	1	-1	0	0	0	0	0
H2	H2	2	0	0	0	-1	0	0	0	0	0
CE	CE	-1	0	0	1	0	0	0	0	0	0
H6	H6	2	0	0	-1	0	-1	0	0	1	0
CA	CA	0	0	0	0	-1	1	0	0	0	0
H3	H3	-2	0	0	0	-1	1	0	0	0	0
D4	D4	-1	0	1	0	0	-1	0	0	0	0
YC	YC	0	0	0	-1	1	0	0	0	0	0
Y2	Y2	2	0	0	-2	-1	0	0	0	0	0
H5	H5	1	0	0	0	0	0	0	0	0	-1
CD	CD	-1	0	-2	0	2	1	0	0	0	-1
EB	EB	0	-1	0	1	0	0	1	0	0	-2
SITE EFFECTS		15	6	3	0	3	0	-7	-9	-13	331

```
BOX PLOT OF  180 STUDENTIZED RESIDUALS FROM LPLT= -2.712    TO ULPT=  2.439
NO.<LPLT                                                          NO.>UPLT
       0 *       -----------------I    +        I--------------------   0

MEDIAN= -0.1267E+00 ANDERSON-DARLING STATISTIC=  0.684

ANALYSIS OF VARIANCE FOR THE ADDITIVE MODEL

-------------------------------------------------------------------
SOURCE                  D.F.     S.S.         M.S.     F       FPROB
-------------------------------------------------------------------
TREATMENTS               19    6741.45        354.813
LOCATIONS                 8    211319.        26414.9
TREATMENT X SITES       152    55261.0        363.559
-------------------------------------------------------------------
TOTAL                   179    273322.
```

RESIDUALS FROM THE ADDITIVE SITE X TREATMENT MODEL FILE 07_ELITE   16/ 8/ 9 12:59
 -- :PAGE   5

 SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
D3	D3	8.663	-19.67	-5.717	-5.714	-7.073	-13.13	-6.416
H1	H1	13.34	-13.97	10.60	-14.81	0.9589	17.48	-6.432
G2	G2	-16.43	42.05 *	29.16	16.68	3.254	-16.45	-2.257
EA	EA	8.931	19.55	-14.34	-.6508	-5.031	-24.74	19.60
D2	D2	10.50	8.297	-29.08	-10.49	4.526	28.50	-6.012
D1	D1	-6.466	39.41 *	-33.94	-1.641	12.12	7.289	-3.400
G1	G1	-28.77	12.16	6.218	-.9395	25.35	-24.90	-.1210
CC	CC	16.51	-17.53	-8.195	-7.263	-3.616	13.90	-8.028
H4	H4	19.96	-27.79	38.83 *	-8.042	-23.45	-13.45	0.6136
H2	H2	-17.32	-19.91	42.53 *	1.950	-8.260	9.802	3.212
CE	CE	12.88	10.38	-28.39	-2.742	15.02	-14.85	-12.02
H6	H6	-5.095	-15.39	40.88 *	-2.848	-3.066	-25.27	8.512
CA	CA	-10.97	-19.47	-9.909	-5.596	16.96	25.41	-7.165
H3	H3	7.720	-29.19	-35.83 *	10.98	4.383	23.99	12.90
D4	D4	17.49	-4.084	-21.56	-6.896	19.50	-23.29	7.293
YC	YC	-2.183	26.75	3.209	-4.392	7.071	17.44	-11.41
Y2	Y2	-3.187	-25.90	38.37 *	12.64	-.9680	2.034	4.991
H5	H5	-12.85	-4.746	26.72	13.19	-3.383	-15.53	-2.621
CD	CD	16.44	38.93 *	-35.01 *	-2.432	-47.52 **	28.45	0.6043
EB	EB	-29.17	0.1386	-14.53	19.02	-6.779	-2.679	8.157
SITE EFFECTS		24.44 ***	12.54 **	62.19 ***	-31.73 ***	14.16 ***	-1.435	-39.27 ***

SECTION 2

VARIETY\SITE		RI 2007		AL 2007		T-EFCTS	
D3	D3	6.321		42.74	*	11.40	
H1	H1	-4.828		-2.332		7.047	
G2	G2	-9.060		-46.93	**	5.681	
EA	EA	-3.615		0.2966		5.136	
D2	D2	-4.366		-1.891		4.261	
D1	D1	-4.499		-8.869		3.212	
G1	G1	-7.832		18.83		3.165	
CC	CC	-.5727		14.79		2.602	
H4	H4	-4.330		17.66		2.557	
H2	H2	5.175		-17.17		1.670	
CE	CE	-.9741		20.70		1.588	
H6	H6	20.00		-17.72		0.4259	
CA	CA	-6.426		17.17		0.1536	
H3	H3	16.09		-11.03		-2.968	
D4	D4	-1.685		13.23		-3.975	
YC	YC	-8.454		-28.03		-5.127	
Y2	Y2	7.652		-35.63	*	-5.242	
H5	H5	4.624		-5.407		-6.862	
CD	CD	-4.657		5.190		-9.031	
EB	EB	1.439		24.40		-15.70	*
SITE EFFECTS		-54.77	***	13.88		471.6	***

FITTED VALUES FROM ADDITIVE SITE X TREATMENT MODEL FILE 07_ELITE   16/ 8/ 9 12:59
 -- :PAGE   6

 SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
D3	D3	507.5	495.6	545.2	451.3	497.2	481.6	443.8
H1	H1	503.1	491.2	540.9	446.9	492.8	477.2	439.4
G2	G2	501.8	489.8	539.5	445.6	491.5	475.9	438.0
EA	EA	501.2	489.3	539.0	445.0	490.9	475.3	437.5
D2	D2	500.3	488.4	538.1	444.2	490.0	474.5	436.6
D1	D1	499.3	487.4	537.0	443.1	489.0	473.4	435.6
G1	G1	499.2	487.3	537.0	443.1	488.9	473.4	435.5
CC	CC	498.7	486.8	536.4	442.5	488.4	472.8	435.0
H4	H4	498.6	486.7	536.4	442.5	488.3	472.7	434.9
H2	H2	497.7	485.8	535.5	441.6	487.5	471.9	434.0
CE	CE	497.7	485.8	535.4	441.5	487.4	471.8	433.9
H6	H6	496.5	484.6	534.2	440.3	486.2	470.6	432.8
CA	CA	496.2	484.3	534.0	440.0	485.9	470.3	432.5
H3	H3	493.1	481.2	530.8	436.9	482.8	467.2	429.4
D4	D4	492.1	480.2	529.8	435.9	481.8	466.2	428.4
YC	YC	490.9	479.0	528.7	434.8	480.7	465.1	427.2
Y2	Y2	490.8	478.9	528.6	434.7	480.5	464.9	427.1
H5	H5	489.2	477.3	527.0	433.0	478.9	463.3	425.5
CD	CD	487.0	475.1	524.8	430.9	476.8	461.2	423.3
EB	EB	480.4	468.5	518.1	424.2	470.1	454.5	416.7
SITE ESTS.		496.1	484.2	533.8	439.9	485.8	470.2	432.4

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	T-ESTS.
D3	D3	428.3	496.9	483.0
H1	H1	423.9	492.6	478.7
G2	G2	422.5	491.2	477.3
EA	EA	422.0	490.6	476.8
D2	D2	421.1	489.8	475.9
D1	D1	420.1	488.7	474.8
G1	G1	420.0	488.7	474.8
CC	CC	419.5	488.1	474.2
H4	H4	419.4	488.1	474.2
H2	H2	418.5	487.2	473.3
CE	CE	418.4	487.1	473.2
H6	H6	417.3	485.9	472.1
CA	CA	417.0	485.7	471.8
H3	H3	413.9	482.5	468.7
D4	D4	412.9	481.5	467.7
YC	YC	411.7	480.4	466.5
Y2	Y2	411.6	480.3	466.4
H5	H5	410.0	478.6	464.8
CD	CD	407.8	476.5	462.6
EB	EB	401.2	469.8	455.9
SITE ESTS.		416.9	485.5	471.6

 REGRESSIONS OF EOLT_ASI FOR EACH VARIETY ON MEANS OF EOLT_ASI AT EACH SITE

VARIETY		MEAN	SLOPE	SE	MS-TXL	MS-REG	MS-DEV	R**2(%)
CA	CA	471.78	1.012	0.162	243.70	1.46	278.30	0.
CC	CC	474.23	1.036	0.126	148.68	14.06	167.91	1.
CE	CE	473.22	0.982	0.169	263.40	3.31	300.55	0.
CD	CD	462.60	0.847	0.282	766.70	248.34	840.75	4.
D1	D1	474.84	0.877	0.198	382.50	161.08	414.13	5.
D2	D2	475.89	0.939	0.164	253.29	39.08	283.89	2.
D3	D3	483.03	1.020	0.189	332.23	4.11	379.11	0.
D4	D4	467.65	0.958	0.163	248.63	18.87	281.45	1.
YC	YC	466.50	1.115	0.162	261.45	140.45	278.74	7.
Y2	Y2	466.39	1.043	0.222	458.89	19.68	521.63	1.
EA	EA	476.76	0.904	0.147	212.82	96.77	229.40	6.
EB	EB	455.93	0.776	0.147	267.64	532.02	229.87	25.
G1	G1	474.79	1.090	0.188	336.70	86.40	372.46	3.
G2	G2	477.31	1.134	0.274	717.16	188.92	792.62	3.
H1	H1	478.67	1.166	0.103	135.15	290.84	112.90	27.
H2	H2	473.30	1.107	0.197	375.62	120.99	412.00	4.
H3	H3	468.66	0.596*	0.153	432.24	1723.39	247.79	50.
H4	H4	474.18	1.280	0.201	475.62	826.34	425.52	22.
H5	H5	464.77	1.059	0.135	173.19	36.25	192.76	3.
H6	H6	472.05	1.060	0.212	422.01	37.79	476.90	1.

 SLOPE - SLOPES OF REGRESSIONS OF VARIETY MEANS ON SITE INDEX.
          * INDICATES SLOPES SIGNIFICANTLY DIFFERENT FROM THE
          SLOPE FOR THE OVERALL REGRESSION WHICH IS   1.00
 MS-TXL - CONTRIBUTION OF EACH VARIETY TO INTERACTION MS
 MS-REG - CONTRIBUTION OF EACH VARIETY TO THE REGRESSION
          COMPONENT OF THE TREATMENT BY LOCATION INTERACTION
 MS-DEV - DEVIATIONS FROM REGRESSION COMPONENT OF INTERACTION
 R**2   - SQUARED CORRELATION BETWEEN RESIDUALS FROM THE MAIN
          EFFECTS MODEL AND THE SITE INDEX.

VARIATE EOLT_ASI WAS SITE INDEX WITH OVERALL MEAN 471.6
THE FOLLOWING SITE MEANS OF EOLT_ASI WERE USED AS X-VARIATES

496.1	484.2	533.8	439.9	485.8	470.2	432.4	416.9	485.5

ANOVA FOR VARIABLE EOLT_ASI WITH SITE REGRESSIONS ON EOLT_ASI

SOURCE	D.F.	S.S.	M.S.	F	FPROB
TREATMENTS	19	6741.45	354.813		
LOCATIONS	8	211319.	26414.9		
TREATMENT X SITES	152	55261.0	363.559		
TRT X SITE REG	19	4590.13	241.586	0.634	0.875
DEVIATIONS	133	50670.9	380.984		
TOTAL	179	273322.			

```
AMMI ANALYSIS  FILE 07_ELITE   16/ 8/ 9 12:59
 ------------------------------------------------------------- :PAGE   8
 SINGULAR VALUES OF INTERACTION MATRIX (CONDITION=  0)
 141.13    114.93    93.761    74.402    59.752    56.542    24.350    21.103

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR TREATMENTS

 CA CA          CA           0.119993E+01 0.196761E+01-0.986543E+00-0.323142E+01
 CC CC          CC           0.118336E+01 0.242451E+01-0.151064E+01 0.258376E+00
 CE CE          CE           0.282257E+01 0.622990E+00 0.250711E+01-0.149625E+00
 CD CD          CD           0.435035E+01-0.209679E+01-0.291475E+01 0.498699E+01
 D1 D1          D1           0.322420E+01-0.353408E+01 0.676723E+00-0.106961E+01
 D2 D2          D2           0.307501E+01-0.529069E+00-0.225864E+01-0.964013E+00
 D3 D3          D3           0.999387E+00 0.410152E+01 0.184351E+01 0.144585E+01
 D4 D4          D4           0.164743E+01 0.139079E+01 0.257780E+01-0.481261E+00
 YC YC          YC           0.114906E+00-0.358735E+01-0.164393E+01-0.676104E+00
 Y2 Y2          Y2          -0.450896E+01-0.223368E+00-0.234464E+01-0.112776E+01
 EA EA          EA           0.106883E+01-0.114521E+01 0.224849E+01 0.207040E+01
 EB EB          EB           0.100547E+01 0.568951E+00 0.190178E+01-0.759458E+00
 G1 G1          G1          -0.446796E+00-0.479794E+00 0.467908E+01-0.186563E+01
 G2 G2          G2          -0.297457E+01-0.611814E+01 0.985131E+00 0.469867E+00
 H1 H1          H1          -0.412712E+00 0.121116E+01-0.224698E+01-0.552501E-01
 H2 H2          H2          -0.409524E+01 0.210661E+00-0.196285E+01-0.523838E+00
 H3 H3          H3           0.185083E+01 0.152946E+01-0.312537E+01-0.302856E+01
 H4 H4          H4          -0.282527E+01 0.354969E+01-0.217718E+00 0.378820E+01
 H5 H5          H5          -0.278664E+01-0.245132E+00 0.107152E+01 0.239157E+00
 H6 H6          H6          -0.449208E+01 0.381594E+00 0.720915E+00 0.673691E+00

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR ENVIRONMENTS

 PI             PI 2007      0.213484E+01 0.227599E+01-0.228031E+01 0.358085E+01
 KL             KL 2007      0.343842E+01-0.809742E+01 0.245552E+01 0.276560E+01
 LA             LA 2007     -0.991111E+01 0.163890E+00-0.142141E+00 0.175647E+01
 ME             ME 2007     -0.112855E+01-0.137103E+01 0.471919E+00-0.924361E+00
 RO             RO 2007      0.232194E+00-0.402944E+00 0.307158E+01-0.654982E+01
 TY             TY 2007      0.286247E+01-0.257595E+00-0.748925E+01-0.204954E+01
 NA             NA 2007     -0.569887E+00 0.980280E-01 0.431250E+00 0.338921E+00
 RI             RI 2007     -0.101431E+01 0.122379E+01-0.552648E+00-0.505495E+00
 AL             AL 2007      0.395594E+01 0.636729E+01 0.403409E+01 0.158738E+01
```

RESIDUALS FROM THE AMMI-2 MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN UNITS OF ROOT (RESIDUAL GXE MS), ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		LA	PI	RO	AL	KL	TY	ME	NA	RI	TE FCTS
		2	2	2	2	2	2	2	2	2	
		0	0	0	0	0	0	0	0	0	
		0	0	0	0	0	0	0	0	0	
		7	7	7	7	7	7	7	7	7	
D3	D3	0	0	0	0	0	-1	0	0	0	EA
H1	H1	0	0	0	0	0	1	0	0	0	EA
G2	G2	0	0	0	0	0	0	0	0	0	EA
EA	EA	0	0	0	0	0	-1	0	1	0	EA
D2	D2	0	0	0	0	0	1	0	0	0	EA
D1	D1	0	0	0	0	0	0	0	0	0	EA
G1	G1	0	-1	1	1	0	-1	0	0	0	EA
CC	CC	0	0	0	0	0	0	0	0	0	EA
H4	H4	0	1	-1	0	0	0	0	0	0	EA
H2	H2	0	0	0	0	0	1	0	0	0	EA
CE	CE	0	0	0	0	0	-1	0	0	0	EA
H6	H6	0	0	0	0	0	0	0	0	1	EA
CA	CA	0	-1	1	0	0	1	0	0	0	EA
H3	H3	-1	0	0	-1	-1	1	1	0	1	EA
D4	D4	0	0	1	0	0	-1	0	0	0	EA
YC	YC	0	0	0	0	0	1	0	0	0	EA
Y2	Y2	0	0	0	-1	0	1	0	0	0	EA
H5	H5	0	0	0	0	0	0	0	0	0	EA
CD	CD	0	0	-3	0	0	1	0	0	0	EA
EB	EB	0	-2	0	1	0	0	1	0	0	EA

```
BOX PLOT OF  180 STANDERSIZED RESIDUALS FROM LPLT= -2.214    TO ULPT=  1.715
NO.<LPLT                                                          NO.>UPLT
       1 *    **  **----------------I     +     I-----------------* *   0
```

ANALYSIS OF VARIANCE FOR THE AMMI MODEL

SOURCE	D.F.	S.S.	M.S.	F	FPROB
TREATMENTS	19	6741.45	354.813		
LOCATIONS	8	211319.	26414.9		
TREATMENT X SITES	152	55261.0	363.559		
AMMI COMPONENT 1	26	19919.0	766.116	2.731	0.000
AMMI COMPONENT 2	24	13209.7	550.404	2.537	0.001
AMMI COMPONENT 3	22	8791.14	399.597	2.396	0.003
AMMI COMPONENT 4	20	5535.59	276.779	2.128	0.013
GXE RESIDUAL	60	7805.57			
TOTAL	179	273322.			

 GENOTYPE MAP SHOWING BEST GENOTYPES OVER THE RANGE OF AMMI-2 SITE SCORES (2 CHRS/PIXEL)

```
  6.37 H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  6.09 H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  5.82 H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  5.55 H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  5.28 H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  5.00 H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  4.73 H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  4.46 H6H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  4.18 H6H6H6H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  3.91 H6H6H6H6H6H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  3.64 H6H6H6H6H6H6H6H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  3.37 H6H6H6H6H6H6H6H6H6H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  3.09 H6H6H6H6H6H6H6H6H6H6H6H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  2.82 H6H6H6H6H6H6H6H6H6H6H6H6H6H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  2.55 H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  2.27 H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H4H4H4H4H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  2.00 H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H4H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  1.73 H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  1.45 H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H4H4H4H4H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  1.18 H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H4H4H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  0.91 H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H4H4D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  0.64 H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H2H2D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  0.36 H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H2H2H2D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3
  0.09 H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6G2G2G2G2G2D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D2D2
 -0.18 H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6G2G2G2G2G2G2G2G2G2G2G2G2D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D3D2D1D1D1
 -0.46 H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2D3D3D3D3D3D3D3D3D3D1D1D1D1D1D1D1D1
 -0.73 H6H6H6H6H6H6H6H6H6H6H6H6G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2D3D3D3D1D1D1D1D1D1D1D1D1D1
 -1.00 H6H6H6H6H6H6H6H6G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2D1D1D1D1D1D1D1D1D1D1D1D1D1
 -1.27 H6H6H6H6G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2D1D1D1D1D1D1D1D1D1D1D1D1D1
 -1.55 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2D1D1D1D1D1D1D1D1D1D1D1D1D1
 -1.82 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2D1D1D1D1D1D1D1D1D1D1D1D1D1
 -2.09 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2D1D1D1D1D1D1D1D1D1D1D1D1D1
 -2.37 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2D1D1D1D1D1D1D1D1D1D1D1D1D1
 -2.64 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2D1D1D1D1D1D1D1D1D1D1D1D1D1
 -2.91 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2D1D1D1D1D1D1D1D1D1D1D1D1D1
 -3.18 G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2D1D1D1D1D1D1D1D1D1D1D1D1D1
```

```
-3.46 G2G2G2
```

AMMI RESIDUALS, ADDITIVE EFFECTS AND MULTIPLICATIVE SCORES FILE 07_ELITE   16/ 8/ 9 12:59
 -- :PAGE 10

 SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
D3	D3	-2.805	10.10	3.516	1.038	-5.652	-14.94	-6.248
H1	H1	11.46	-2.739	6.307	-13.62	1.543	18.97	-6.786
G2	G2	3.841	2.735	0.6777	4.930	1.480	-9.513	-3.352
EA	EA	9.256	6.598	-3.561	-1.015	-5.740	-28.09	20.32
D2	D2	5.143	-6.560	1.486	-7.740	3.599	19.57	-4.208
D1	D1	-5.306	-.2977	-1.408	-2.848	9.951	-2.851	-1.216
G1	G1	-26.72	9.812	1.869	-2.102	25.26	-23.74	-.3286
CC	CC	8.470	-1.962	3.136	-2.603	-2.913	11.14	-7.592
H4	H4	17.91	10.67	10.25	-6.364	-21.36	-4.453	-1.344
H2	H2	-9.060	-4.123	1.902	-2.383	-7.225	21.58	0.8574
CE	CE	5.437	5.718	-.5168	1.297	14.61	-22.77	-10.47
H6	H6	3.626	3.143	-3.709	-7.394	-1.870	-12.31	5.914
CA	CA	-18.01	-7.665	1.662	-1.544	17.47	22.48	-6.674
H3	H3	0.2875	-23.17	-17.74	15.17	4.570	19.08	13.80
D4	D4	10.81	1.513	-5.460	-3.130	19.68	-27.65	8.096
YC	YC	5.737	-2.696	4.936	-9.181	5.599	16.19	-10.99
Y2	Y2	6.947	-12.21	-6.284	7.248	-.1108E-01	14.88	2.443
H5	H5	-6.341	2.851	-.8561	9.704	-2.835	-7.614	-4.185
CD	CD	11.93	6.994	8.452	-.3975	-49.37 ***	15.45	3.289
EB	EB	-32.61 *	1.288	-4.656	20.93	-6.784	-5.411	8.674
SITE EFFECTS		24.44 ***	12.54	62.19	-31.73	14.16 ***	-1.435 ***	-39.27
AMMI1 SITE		2.135	3.438 ***	-9.911	-1.129	0.2322	2.862 ***	-.5699 ***
AMMI2 SITE		2.276 ***	-8.097 ***	0.1639	-1.371	-.4029 ***	-.2576	0.9803E-01

SECTION 2

VARIETY\SITE		RI 2007		AL 2007		T-EFCTS		AMMI1 TRT		AMMI2 TRT	
D3	D3	2.315		12.67		11.40	***	0.9994	***	4.102	***
H1	H1	-6.729		-8.411		7.047	***	-.4127	***	1.211	
G2	G2	-4.590		3.792		5.681	***	-2.975	***	-6.118	***
EA	EA	-1.129		3.360		5.136	***	1.069		-1.145	
D2	D2	-.5991		-10.69		4.261		3.075	***	-.5291	***
D1	D1	3.096		0.8790		3.212	***	3.224	***	-3.534	
G1	G1	-7.698		23.65		3.165	***	-.4468	***	-.4798	***
CC	CC	-2.339		-5.331		2.602	***	1.183		2.425	
H4	H4	-11.54		6.237		2.557	***	-2.825	***	3.550	
H2	H2	0.7637		-2.311		1.670	***	-4.095		0.2107	***
CE	CE	1.126		5.569		1.588	***	2.823		0.6230	***
H6	H6	14.98		-2.381		0.4259		-4.492		0.3816	***
CA	CA	-7.617		-.1030		0.1536		1.200	***	1.968	***
H3	H3	16.09		-28.09		-2.968	***	1.851		1.529	***
D4	D4	-1.716		-2.141		-3.975		1.647		1.391	***
YC	YC	-3.947		-5.646		-5.127		0.1149	***	-3.587	***
Y2	Y2	3.351		-16.37		-5.242		-4.509		-.2234	
H5	H5	2.097		7.178		-6.862	***	-2.787		-.2451	
CD	CD	2.322		1.331		-9.031	***	4.350		-2.097	
EB	EB	1.762		16.80		-15.70		1.005		0.5690	
SITE EFFECTS		-54.77	***	13.88	***	471.6	***	.		.	
AMMI1 SITE		-1.014		3.956		.		.		.	
AMMI2 SITE		1.224	***	6.367		.		.		.	

FITTED VALUES FROM THE AMMI MODEL FILE 07_ELITE   16/ 8/ 9 12:59
---------- :PAGE 11

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
D3	D3	518.9	465.8	536.0	444.5	495.8	483.4	443.6
H1	H1	505.0	480.0	545.2	445.7	492.2	475.7	439.8
G2	G2	481.5	529.2	568.0	457.3	493.2	468.9	439.1
EA	EA	500.9	502.3	528.2	445.4	491.6	478.7	436.8
D2	D2	505.7	503.3	507.5	441.4	491.0	483.4	434.8
D1	D1	498.1	527.1	504.5	444.3	491.2	483.5	433.4
G1	G1	497.2	489.7	541.3	444.2	489.0	472.2	435.7
CC	CC	506.7	471.2	525.1	437.8	487.7	475.6	434.5
H4	H4	500.7	448.3	565.0	440.8	486.3	463.7	436.9
H2	H2	489.5	470.1	576.1	445.9	486.4	460.1	436.4
CE	CE	505.1	490.4	507.5	437.4	487.8	479.7	432.4
H6	H6	487.8	466.1	578.8	444.9	485.0	457.7	435.4
CA	CA	503.3	472.5	522.4	436.0	485.4	473.3	432.0
H3	H3	500.5	475.2	512.8	432.7	482.6	472.1	428.5
D4	D4	498.8	474.6	513.7	432.2	481.6	470.6	427.6
YC	YC	483.0	508.5	527.0	439.6	482.1	466.3	426.8
Y2	Y2	480.7	465.2	573.2	440.0	479.6	452.1	429.7
H5	H5	482.7	469.7	554.5	436.5	478.4	455.4	427.1
CD	CD	491.6	507.1	481.3	428.8	478.6	474.2	420.6
EB	EB	483.8	467.3	508.2	422.3	470.1	457.2	416.1
SITE ESTS.		496.1	484.2	533.8	439.9	485.8	470.2	432.4
AMMI1 SITE		2.135	3.438	-9.911	-1.129	0.2322	2.862	-.5699
AMMI2 SITE		2.276	-8.097	0.1639	-1.371	-.4029	-.2576	0.9803E-01

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	T-ESTS.	AMMI1 TRT	AMMI2 TRT
D3	D3	432.3	527.0	483.0	0.9994	4.102 I
H1	H1	425.8	498.6	478.7	-.4127	1.211 I
G2	G2	418.1	440.5	477.3	-2.975	-6.118 I
EA	EA	419.5	487.6	476.8	1.069	-1.145 I
D2	D2	417.4	498.6	475.9	3.075	-.5291 I
D1	D1	412.5	479.0	474.8	3.224	-3.534 I
G1	G1	419.9	483.8	474.8	-.4468	-.4798 I
CC	CC	421.2	508.2	474.2	1.183	2.425 I
H4	H4	426.6	499.5	474.2	-2.825	3.550 I
H2	H2	422.9	472.3	473.3	-4.095	0.2107 I
CE	CE	416.3	502.2	473.2	2.823	0.6230 I
H6	H6	422.3	470.6	472.1	-4.492	0.3816 I
CA	CA	418.2	502.9	471.8	1.200	1.968 I
H3	H3	413.9	499.6	468.7	1.851	1.529 I
D4	D4	412.9	496.9	467.7	1.647	1.391 I
YC	YC	407.2	458.0	466.5	0.1149	-3.587 I
Y2	Y2	415.9	461.0	466.4	-4.509	-.2234 I
H5	H5	412.5	466.1	464.8	-2.787	-.2451 I
CD	CD	400.8	480.3	462.6	4.350	-2.097 I
EB	EB	400.8	477.4	455.9	1.005	0.5690 I
SITE ESTS.		416.9	485.5	471.6	.	.
AMMI1 SITE		-1.014	3.956	.	.	.
AMMI2 SITE		1.224 I	6.367 I	.	.	.

Addendum 10: Cross-site analysis of relative ethanol output (REO), the 2007 season trials

```
PBGXE - CROSS SITE ANALYSIS  FILE 07_ELITE   17/ 8/ 9 21:27
 ------------------------------------------------------------------ :PAGE   1

  20       CN$    CODES:
  1 CA          CA            2 CC          CC            3 CE          CE
  4 CD          CD            5 D1          D1            6 D2          D2
  7 D3          D3            8 D4          D4            9 YC          YC
 10 Y2          Y2           11 EA          EA           12 EB          EB
 13 G1          G1           14 G2          G2           15 H1          H1
 16 H2          H2           17 H3          H3           18 H4          H4
 19 H5          H5           20 H6          H6

   9     SITE$    CODES:
  1 PI 2007       2 KL 2007       3 LA 2007       4 ME 2007       5 RO 2007
  6 TY 2007       7 NA 2007       8 RI 2007       9 AL 2007

 TREATMENT BY VARIATE MEANS HAVE BEEN REQUESTED FOR  1 VARIATES:
 EO_%_TH
 ROWS OF MEANS TABLES TO BE SORTED ON VARIATE EO_%_TH
 GXE ANALYSIS HAS BEEN REQUESTED FOR  1 VARIATES
```

TREATMENT MEANS AND COUNTS OVER SITES FOR EACH VARIATE. FILE 07_ELITE 17/ 8/ 9 21:27
:PAGE 2

VARIETY\SITE		EO_%_TH	
CC	CC	105.0	27
H1	H1	104.1	27
D3	D3	103.9	27
CE	CE	103.9	27
D1	D1	103.7	27
YC	YC	103.7	27
EA	EA	103.6	27
CA	CA	103.1	27
D2	D2	103.1	27
H4	H4	103.0	27
G2	G2	102.8	27
H2	H2	102.3	27
Y2	Y2	102.2	27
H5	H5	101.9	27
CD	CD	101.3	27
EB	EB	101.3	27
D4	D4	101.2	27
G1	G1	100.9	27
H6	H6	100.0	27
H3	H3	99.74	27
SITE MEANS		102.5	540

20 X 9 MATRIX OF TREATMENT BY SITE MEANS FOR VARIATE EO_%_TH FILE 07_ELITE 17/ 8/ 9 21:27
:PAGE 3

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
CC	CC	110.1	99.24	112.6	95.57	108.2	112.8	93.48
H1	H1	107.6	104.1	115.7	91.05	105.0	109.7	96.98
D3	D3	109.6	102.3	115.5	95.62	102.3	104.6	90.04
CE	CE	107.2	108.5	110.1	95.16	103.9	103.0	91.88
D1	D1	101.6	116.1	108.9	96.27	107.2	106.4	94.06
YC	YC	102.3	113.1	114.9	97.60	105.4	110.7	89.30
EA	EA	107.1	108.4	110.5	92.66	102.3	102.5	97.49
CA	CA	99.75	105.3	110.0	91.71	109.0	109.6	94.24
D2	D2	109.2	106.7	109.9	92.52	102.4	111.9	92.61
H4	H4	111.4	97.28	122.7	93.87	100.9	102.7	91.01
G2	G2	100.8	115.8	119.6	98.96	106.3	102.1	92.81
H2	H2	102.3	101.4	121.5	94.95	102.2	105.5	92.85
Y2	Y2	104.2	101.7	118.8	95.75	102.3	105.2	94.71
H5	H5	98.68	100.6	116.4	97.20	101.4	104.2	94.73
CD	CD	105.4	112.0	105.7	95.04	89.73	107.8	94.03
EB	EB	101.1	100.3	107.9	96.64	98.81	106.7	93.25
D4	D4	108.7	100.9	108.5	92.95	103.5	98.42	89.74
G1	G1	96.28	105.5	113.7	91.99	106.2	98.81	91.63
H6	H6	102.4	99.45	120.0	89.98	103.2	99.60	91.94
H3	H3	102.7	97.83	102.4	94.36	102.5	105.4	96.09
SITE MEANS		104.4	104.8	113.3	94.49	103.1	105.4	93.14
SITE INDEX		104.4	104.8	113.3	94.49	103.1	105.4	93.14

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	TRT MEANS
CC	CC	97.27	115.8	105.0
H1	H1	97.46	109.5	104.1
D3	D3	95.12	120.6	103.9
CE	CE	94.91	120.1	103.9
D1	D1	95.34	107.4	103.7
YC	YC	95.73	104.0	103.7
EA	EA	97.56	114.1	103.6
CA	CA	95.69	113.0	103.1
D2	D2	93.05	110.0	103.1
H4	H4	93.82	113.1	103.0
G2	G2	92.05	96.59	102.8
H2	H2	95.83	104.2	102.3
Y2	Y2	94.34	102.5	102.2
H5	H5	97.50	106.6	101.9
CD	CD	94.91	107.0	101.3
EB	EB	92.85	113.9	101.3
D4	D4	96.37	112.1	101.2
G1	G1	92.89	111.4	100.9
H6	H6	92.36	101.5	100.0
H3	H3	92.79	103.6	99.74
SITE MEANS		94.89	109.3	102.5
SITE INDEX		94.89	109.3	102.5

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

ENVIRONMENT	MEAN	SE	DUNCAN GROUPS	LSD TESTS
LA 2007	113.27	0.97246	.	
AL 2007	109.34	0.97246	2.	
TY 2007	105.37	0.97246	| 32.	
KL 2007	104.83	0.97246	| 32..	
PI 2007	104.42	0.97246	| 33...	
RO 2007	103.14	0.97246	| 33....	
RI 2007	94.892	0.97246	| 333333.	
ME 2007	94.492	0.97246	| 333333..	
NA 2007	93.142	0.97246	| 333333...	

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

TREATMENT		MEAN	SE	DUNCAN GROUPS	LSD TESTS
CC	CC	105.00	1.4497	| .	
H1	H1	104.12	1.4497	|| ..	
D3	D3	103.95	1.4497	|| ...	
CE	CE	103.86	1.4497	||	
D1	D1	103.70	1.4497	||	
YC	YC	103.68	1.4497	||	
EA	EA	103.63	1.4497	||	
CA	CA	103.14	1.4497	||	
D2	D2	103.12	1.4497	||	
H4	H4	102.97	1.4497	||	
G2	G2	102.78	1.4497	||	
H2	H2	102.31	1.4497	||	
Y2	Y2	102.16	1.4497	||	
H5	H5	101.92	1.4497	||	
CD	CD	101.29	1.4497	||	
EB	EB	101.27	1.4497	||	
D4	D4	101.25	1.4497	||	
G1	G1	100.94	1.4497	|| 1.....	
H6	H6	100.05	1.4497	|| 11.....	
H3	H3	99.740	1.4497	| 1111.....	

RESIDUALS FROM THE ADDITIVE TREATMENT BY SITE MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN STANDARD ERRORS,
 ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		LA	AL	TY	KL	PI	RO	RI	ME	NA		T-E FCTS
		2	2	2	2	2	2	2	2	2		
		0	0	0	0	0	0	0	0	0		
		0	0	0	0	0	0	0	0	0		
		7	7	7	7	7	7	7	7	7		
CC	CC	0	0	1	-2	0	0	0	0	0		1
H1	H1	0	0	0	0	0	0	0	-1	0		1
D3	D3	0	2	0	0	0	0	0	0	-1		0
CE	CE	-1	2	0	0	0	0	0	0	0		0
D1	D1	-1	0	0	2	-1	0	0	0	0		0
YC	YC	0	-1	1	1	0	0	0	0	-1		0
EA	EA	0	0	0	0	0	0	0	0	0		0
CA	CA	0	0	0	0	-1	1	0	0	0		0
D2	D2	0	0	1	0	1	0	0	0	0		0
H4	H4	2	0	0	-1	1	0	0	0	0		0
G2	G2	1	-3	0	2	0	0	0	1	0		0
H2	H2	2	-1	0	0	0	0	0	0	0		0
Y2	Y2	1	-1	0	0	0	0	0	0	0		0
H5	H5	0	0	0	0	-1	0	0	0	0		0
CD	CD	-1	0	0	2	0	-3	0	0	0		0
EB	EB	-1	1	0	0	0	0	0	0	0		0
D4	D4	0	1	-1	0	1	0	0	0	0		0
G1	G1	0	0	-1	0	-1	1	0	0	0		-1
H6	H6	2	-1	0	0	0	0	0	0	0		-2
H3	H3	-2	0	0	-1	0	0	0	0	1		-1
SITE EFFECTS		11	0	3	2	2	0	-8	-8	-10		316

```
BOX PLOT OF  180 STUDENTIZED RESIDUALS FROM LPLT= -3.249    TO ULPT=  2.691
NO.<LPLT                                                          NO.>UPLT
       0 * *         ------------I      +      I--------------------   0

MEDIAN= -0.2433E-01 ANDERSON-DARLING STATISTIC=  0.460

ANALYSIS OF VARIANCE FOR THE ADDITIVE MODEL

-------------------------------------------------------------------
SOURCE                  D.F.     S.S.         M.S.     F       FPROB
-------------------------------------------------------------------
TREATMENTS               19    351.861        18.5190
LOCATIONS                 8    7800.39        975.048
TREATMENT X SITES       152    2874.86        18.9136
-----------------------------------------------------------------
TOTAL                   179    11027.1
```

RESIDUALS FROM THE ADDITIVE SITE X TREATMENT MODEL FILE 07_ELITE   17/ 8/ 9 21:27
 -- :PAGE   5

 SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
CC	CC	3.222	-8.048 *	-3.087	-1.384	2.569	4.979	-2.119
H1	H1	1.655	-2.334	0.8822	-5.013	0.2524	2.736	2.262
D3	D3	3.761	-3.981	0.7902	-.2818	-2.287	-2.196	-4.503
CE	CE	1.500	2.315	-4.472	-.6439	-.5367	-3.726	-2.579
D1	D1	-4.009	10.15 *	-5.482	0.6242	2.887	-.1675	-.2409
YC	YC	-3.246	7.189	0.5516	1.974	1.119	4.160	-4.978
EA	EA	1.608	2.523	-3.878	-2.925	-1.923	-3.916	3.260
CA	CA	-5.266	-.1119	-3.864	-3.379	5.303	3.586	0.4952
D2	D2	4.162	1.281	-3.979	-2.540	-1.348	5.905	-1.108
H4	H4	6.506	-7.971 *	9.002 *	-1.052	-2.625	-3.125	-2.557
G2	G2	-3.876	10.75 **	6.097	4.235	2.954	-3.546	-.5613
H2	H2	-1.859	-3.200	8.468 *	0.6952	-.6734	0.3881	-.6343E-01
Y2	Y2	0.1411	-2.724	5.878	1.636	-.4835	0.2038	1.948
H5	H5	-5.115	-3.648	3.779	3.328	-1.101	-.5298	2.208
CD	CD	2.256	8.406 *	-6.315	1.802	-12.15 **	3.680	2.142
EB	EB	-2.044	-3.248	-4.125	3.420	-3.057	2.640	1.374
D4	D4	5.583	-2.610	-3.488	-.2458	1.662	-5.659	-2.110
G1	G1	-6.535	2.324	2.045	-.8981	4.706	-4.957	0.9143E-01
H6	H6	0.4825	-2.876	9.237 *	-2.019	2.546	-3.281	1.292
H3	H3	1.075	-4.195	-8.040 *	2.668	2.187	2.826	5.747
SITE EFFECTS		1.875 *	2.282 *	10.72 ***	-8.051 ***	0.5980	2.829 **	-9.402 ***

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	T-EFCTS
CC	CC	-.8252E-01	3.951	2.459
H1	H1	0.9944	-1.435	1.575
D3	D3	-1.177	9.875 *	1.406
CE	CE	-1.292	9.436 *	1.315
D1	D1	-.7136	-3.053	1.158
YC	YC	-.2977	-6.471	1.132
EA	EA	1.579	3.672	1.089
CA	CA	0.2008	3.035	0.6002
D2	D2	-2.418	0.4515E-01	0.5728
H4	H4	-1.502	3.325	0.4287
G2	G2	-3.071	-12.99 **	0.2321
H2	H2	1.172	-4.928	-.2329
Y2	Y2	-.1670	-6.433	-.3829
H5	H5	3.235	-2.156	-.6250
CD	CD	1.274	-1.094	-1.256
EB	EB	-.7749	5.814	-1.270
D4	D4	2.770	4.099	-1.296
G1	G1	-.3942	3.618	-1.606
H6	H6	-.4039E-01	-5.341	-2.496 **
H3	H3	0.7056	-2.974	-2.803 *
SITE EFFECTS		-7.651 ***	6.799	102.5 ***

FITTED VALUES FROM ADDITIVE SITE X TREATMENT MODEL FILE 07_ELITE   17/ 8/ 9 21:27
---------- :PAGE   6

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
CC	CC	106.9	107.3	115.7	96.95	105.6	107.8	95.60
H1	H1	106.0	106.4	114.8	96.07	104.7	106.9	94.72
D3	D3	105.8	106.2	114.7	95.90	104.5	106.8	94.55
CE	CE	105.7	106.1	114.6	95.81	104.5	106.7	94.46
D1	D1	105.6	106.0	114.4	95.65	104.3	106.5	94.30
YC	YC	105.6	106.0	114.4	95.62	104.3	106.5	94.27
EA	EA	105.5	105.9	114.4	95.58	104.2	106.5	94.23
CA	CA	105.0	105.4	113.9	95.09	103.7	106.0	93.74
D2	D2	105.0	105.4	113.8	95.07	103.7	105.9	93.71
H4	H4	104.8	105.3	113.7	94.92	103.6	105.8	93.57
G2	G2	104.7	105.1	113.5	94.72	103.4	105.6	93.37
H2	H2	104.2	104.6	113.0	94.26	102.9	105.1	92.91
Y2	Y2	104.0	104.4	112.9	94.11	102.8	105.0	92.76
H5	H5	103.8	104.2	112.6	93.87	102.5	104.7	92.52
CD	CD	103.2	103.6	112.0	93.24	101.9	104.1	91.89
EB	EB	103.1	103.6	112.0	93.22	101.9	104.1	91.87
D4	D4	103.1	103.5	112.0	93.20	101.8	104.1	91.85
G1	G1	102.8	103.2	111.7	92.89	101.5	103.8	91.54
H6	H6	101.9	102.3	110.8	92.00	100.6	102.9	90.65
H3	H3	101.6	102.0	110.5	91.69	100.3	102.6	90.34
SITE ESTS.		104.4	104.8	113.3	94.49	103.1	105.4	93.14

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	T-ESTS.
CC	CC	97.35	111.8	105.0
H1	H1	96.47	110.9	104.1
D3	D3	96.30	110.7	103.9
CE	CE	96.21	110.7	103.9
D1	D1	96.05	110.5	103.7
YC	YC	96.02	110.5	103.7
EA	EA	95.98	110.4	103.6
CA	CA	95.49	109.9	103.1
D2	D2	95.46	109.9	103.1
H4	H4	95.32	109.8	103.0
G2	G2	95.12	109.6	102.8
H2	H2	94.66	109.1	102.3
Y2	Y2	94.51	109.0	102.2
H5	H5	94.27	108.7	101.9
CD	CD	93.64	108.1	101.3
EB	EB	93.62	108.1	101.3
D4	D4	93.60	108.0	101.2
G1	G1	93.29	107.7	100.9
H6	H6	92.40	106.8	100.0
H3	H3	92.09	106.5	99.74
SITE ESTS.		94.89	109.3	102.5

REGRESSIONS OF EO_%_TH FOR EACH VARIETY ON MEANS OF EO_%_TH AT EACH SITE

VARIETY		MEAN	SLOPE	SE	MS-TXL	MS-REG	MS-DEV	R**2(%)
CA	CA	103.14	1.009	0.196	13.07	0.03	14.93	0.
CC	CC	105.00	1.074	0.223	17.26	2.12	19.43	2.
CE	CE	103.86	1.135	0.220	17.44	7.14	18.92	5.
CD	CD	101.29	0.762	0.312	35.93	22.17	37.90	8.
D1	D1	103.70	0.846	0.241	20.98	9.21	22.67	5.
D2	D2	103.12	1.086	0.173	10.63	2.90	11.73	3.
D3	D3	103.95	1.307	0.212	19.99	36.68	17.61	23.
D4	D4	101.25	0.950	0.203	14.24	0.97	16.14	1.
YC	YC	103.68	1.046	0.235	18.97	0.82	21.56	1.
Y2	Y2	102.16	0.957	0.181	11.27	0.70	12.78	1.
EA	EA	103.63	0.899	0.165	9.77	3.95	10.60	5.
EB	EB	101.27	0.885	0.182	12.01	5.15	12.99	5.
G1	G1	100.94	1.097	0.200	14.13	3.65	15.63	3.
G2	G2	102.78	0.951	0.375	48.18	0.95	54.93	0.
H1	H1	104.12	1.043	0.134	6.23	0.73	7.01	1.
H2	H2	102.31	1.085	0.200	14.02	2.83	15.62	3.
H3	H3	99.74	0.524*	0.145	18.21	88.31	8.19	61.
H4	H4	102.97	1.376	0.249	28.06	55.19	24.19	25.
H5	H5	101.92	0.829	0.166	10.79	11.35	10.71	13.
H6	H6	100.05	1.138	0.225	18.17	7.40	19.71	5.

SLOPE - SLOPES OF REGRESSIONS OF VARIETY MEANS ON SITE INDEX.
         * INDICATES SLOPES SIGNIFICANTLY DIFFERENT FROM THE
         SLOPE FOR THE OVERALL REGRESSION WHICH IS   1.00
MS-TXL - CONTRIBUTION OF EACH VARIETY TO INTERACTION MS
MS-REG - CONTRIBUTION OF EACH VARIETY TO THE REGRESSION
         COMPONENT OF THE TREATMENT BY LOCATION INTERACTION
MS-DEV - DEVIATIONS FROM REGRESSION COMPONENT OF INTERACTION
R**2   - SQUARED CORRELATION BETWEEN RESIDUALS FROM THE MAIN
         EFFECTS MODEL AND THE SITE INDEX.

```
VARIATE EO_%_TH WAS SITE INDEX WITH OVERALL MEAN  102.5
THE FOLLOWING SITE MEANS OF EO_%_TH WERE USED AS X-VARIATES
          104.4     104.8     113.3     94.49     103.1     105.4     93.14     94.89     109.3
-------------------------------------------------------------------
ANOVA FOR VARIABLE EO_%_TH WITH SITE REGRESSIONS ON EO_%_TH
-------------------------------------------------------------------
SOURCE              D.F.      S.S.         M.S.     F       FPROB
-------------------------------------------------------------------
TREATMENTS           19    351.861        18.5190
LOCATIONS             8    7800.39        975.048
TREATMENT X SITES   152    2874.86        18.9136
   TRT X SITE REG    19    262.241        13.8022       0.703   0.811
   DEVIATIONS       133    2612.62        19.6438
-------------------------------------------------------------------
TOTAL               179    11027.1
```

```
AMMI ANALYSIS  FILE 07_ELITE   17/ 8/ 9 21:27
 ------------------------------------------------------------ :PAGE   8
 SINGULAR VALUES OF INTERACTION MATRIX (CONDITION=  0)
 31.587    27.480    18.995    17.821    13.085    12.078    9.6313    5.8124

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR TREATMENTS

 CA CA          CA           0.305504E+00-0.701151E+00 0.524774E+00-0.195805E+01
 CC CC          CC           0.156637E+01 0.356431E+00-0.837976E+00-0.117029E+01
 CE CE          CE           0.140131E+01-0.901309E+00 0.155998E+01 0.659196E+00
 CD CD          CD          -0.182402E+00-0.224331E+01-0.156438E+01 0.223163E+01
 D1 D1          D1          -0.125754E+01-0.189087E+01 0.889791E+00-0.361423E+00
 D2 D2          D2           0.457212E+00-0.860401E+00-0.106012E+01 0.161593E+00
 D3 D3          D3           0.187581E+01 0.520476E+00 0.846485E+00 0.966988E+00
 D4 D4          D4           0.126425E+01 0.111374E+00 0.782832E+00 0.350036E+00
 YC YC          YC          -0.168635E+01-0.780562E+00-0.828256E-01 0.300200E-01
 Y2 Y2          Y2          -0.976872E+00 0.123045E+01-0.913607E+00 0.720837E-01
 EA EA          EA           0.656312E+00-0.777025E+00 0.567428E+00 0.540616E+00
 EB EB          EB           0.110545E+01-0.596011E+00-0.445956E+00-0.314802E+00
 G1 G1          G1          -0.297460E+00 0.128300E+00 0.214589E+01-0.773694E+00
 G2 G2          G2          -0.335175E+01-0.175699E-04 0.731920E+00 0.612572E+00
 H1 H1          H1           0.172197E+00 0.378677E+00-0.816936E+00-0.431389E+00
 H2 H2          H2          -0.943110E+00 0.160234E+01-0.533221E+00 0.743844E-01
 H3 H3          H3           0.320939E+00-0.730168E+00-0.151525E+01-0.153731E+01
 H4 H4          H4           0.106535E+01 0.235264E+01-0.542228E-01 0.134703E+01
 H5 H5          H5          -0.539193E+00 0.843729E+00-0.350047E+00-0.497574E+00
 H6 H6          H6          -0.956022E+00 0.195641E+01 0.125446E+00-0.161484E-02

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR ENVIRONMENTS

 PI             PI 2007      0.180032E+01 0.569250E+00-0.118276E+01 0.186216E+01
 KL             KL 2007     -0.267586E+01-0.311121E+01 0.149301E+01 0.152591E+01
 LA             LA 2007     -0.175700E+01 0.394345E+01 0.532093E+00 0.110286E+01
 ME             ME 2007     -0.818601E+00-0.237046E+00-0.366909E+00 0.375429E+00
 RO             RO 2007     -0.631337E+00 0.632566E+00 0.172317E+01-0.292778E+01
 TY             TY 2007      0.707076E-01-0.105395E+01-0.265858E+01-0.116517E+01
 NA             NA 2007     -0.295752E+00-0.250212E+00-0.111506E+01-0.788434E+00
 RI             RI 2007      0.196789E+00 0.484352E-01-0.314387E+00-0.233705E+00
 AL             AL 2007      0.411074E+01-0.541283E+00 0.188942E+01 0.248733E+00
```

RESIDUALS FROM THE AMMI-2 MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN UNITS OF ROOT (RESIDUAL GXE MS), ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		LA	AL	TY	KL	PI	RO	RI	ME	NA	T-EFCTS
		2	2	2	2	2	2	2	2	2	
		0	0	0	0	0	0	0	0	0	
		0	0	0	0	0	0	0	0	0	
		7	7	7	7	7	7	7	7	7	
CC	CC	0	0	1	0	0	1	0	0	0	EA
H1	H1	0	0	0	0	0	0	0	-1	0	EA
D3	D3	0	0	0	0	0	0	0	0	-1	EA
CE	CE	0	0	-1	0	0	0	0	0	0	EA
D1	D1	0	0	0	0	0	0	0	0	0	EA
YC	YC	0	0	1	0	0	0	0	0	-1	EA
EA	EA	0	0	-1	0	0	0	0	0	0	EA
CA	CA	0	0	0	0	-1	1	0	0	0	EA
D2	D2	0	0	1	0	1	0	0	0	0	EA
H4	H4	0	0	0	0	0	-1	0	0	0	EA
G2	G2	0	0	0	0	0	0	0	0	0	EA
H2	H2	0	0	0	0	0	0	0	0	0	EA
Y2	Y2	0	0	0	0	0	0	0	0	0	EA
H5	H5	0	0	0	0	-1	0	0	0	0	EA
CD	CD	0	0	0	0	1	-3	0	0	0	EA
EB	EB	0	0	0	0	-1	0	0	1	0	EA
D4	D4	0	0	-1	0	0	0	0	0	0	EA
G1	G1	0	1	-1	0	-1	1	0	0	0	EA
H6	H6	0	0	0	0	0	0	0	0	0	EA
H3	H3	-1	-1	0	-1	0	0	0	0	1	EA

```
BOX PLOT OF  180 STANDERSIZED RESIDUALS FROM LPLT= -1.831    TO ULPT=  1.791
NO.<LPLT                                                          NO.>UPLT
       1 ----------------------I        +     I----------------------   0
```

ANALYSIS OF VARIANCE FOR THE AMMI MODEL

SOURCE	D.F.	S.S.	M.S.	F	FPROB
TREATMENTS	19	351.861	18.5190		
LOCATIONS	8	7800.39	975.048		
TREATMENT X SITES	152	2874.86	18.9136		
AMMI COMPONENT 1	26	997.708	38.3734	2.576	0.000
AMMI COMPONENT 2	24	755.127	31.4636	2.860	0.000
AMMI COMPONENT 3	22	360.817	16.4008	1.724	0.042
AMMI COMPONENT 4	20	317.585	15.8793	2.148	0.012
GXE RESIDUAL	60	443.627			
TOTAL	179	11027.1			

 GENOTYPE MAP SHOWING BEST GENOTYPES OVER THE RANGE OF AMMI-2 SITE SCORES (2 CHRS/PIXEL)

```
     3.94 G2G2H2H2H2H2H2H4H4H4
```

```
-0.85 G2G2G2G2G2G2G2G2G2G2G2D1D1D1D1D1D1D1D1D1D1CCCC
```

 SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
CC	CC	0.1992	-2.748	-1.741	-.1730E-01	3.333	5.243	-1.566
H1	H1	1.129	-.6949	-.3085	-4.782	0.1216	3.123	2.408
D3	D3	0.8784E-01	2.658	2.033	1.377	-1.432	-1.781	-3.818
CE	CE	-.5099	3.260	1.544	0.2896	0.9182	-4.775	-2.390
D1	D1	-.6687	0.9067	-.2348	-.8534	3.289	-2.071	-1.086
YC	YC	0.2341	0.2479	0.6668	0.4081	0.5481	3.457	-5.672
EA	EA	0.8684	1.862	0.3388	-2.572	-1.017	-4.781	3.260
CA	CA	-5.417	-1.476	-.5621	-3.295	5.940	2.826	0.4101
D2	D2	3.829	-.1726	0.2178	-2.370	-.5152	4.966	-1.188
H4	H4	3.249	2.199	1.596	0.3775	-3.441	-.7208	-1.653
G2	G2	2.158	1.785	0.2077	1.491	0.8384	-3.309	-1.553
H2	H2	-1.073	-.7381	0.4926	0.3030	-2.282	2.144	0.5857E-01
Y2	Y2	1.199	-1.510	-.6906	1.128	-1.879	1.570	1.967
H5	H5	-4.625	-2.466	-.4955	3.087	-1.975	0.3976	2.260
CD	CD	3.861	0.9387	2.211	1.121	-10.85 ***	1.328	1.526
EB	EB	-3.695	-2.145	0.1680	4.184	-1.982	1.934	1.552
D4	D4	3.243	1.119	-1.706	0.8155	2.390	-5.631	-1.708
G1	G1	-6.073	1.927	1.016	-1.111	4.437	-4.800	0.3556E-01
H6	H6	1.090	0.6528	-.1576	-2.338	0.7048	-1.152	1.499
H3	H3	0.9131	-5.608	-4.597	2.757	2.852	2.033	5.659
SITE EFFECTS		1.875 ***	2.282 ***	10.72	-8.051 ***	0.5980	2.829 ***	-9.402 ***
AMMI1 SITE		1.800	-2.676 ***	-1.757	-.8186 ***	-.6313	0.7071E-01	-.2958 ***
AMMI2 SITE		0.5692 ***	-3.111 ***	3.943	-.2370	0.6326	-1.054 ***	-.2502 ***

SECTION 2

VARIETY\SITE		RI 2007		AL 2007		T-EFCTS		AMMI1 TRT		AMMI2 TRT	
CC	CC	-.4080		-2.295		2.459	***	1.566		0.3564	***
H1	H1	0.9422		-1.938		1.575	***	0.1722	***	0.3787	***
D3	D3	-1.571		2.446		1.406		1.876	***	0.5205	***
CE	CE	-1.525		3.188		1.315	***	1.401		-.9013	
D1	D1	-.3746		1.093		1.158	***	-1.258	***	-1.891	***
YC	YC	0.7200E-01		0.3829E-01		1.132		-1.686		-.7806	***
EA	EA	1.487		0.5537		1.089		0.6563		-.7770	
CA	CA	0.1747		1.400		0.6002		0.3055		-.7012	
D2	D2	-2.467		-2.300		0.5728	***	0.4572	***	-.8604	
H4	H4	-1.825		0.2186		0.4287	***	1.065	***	2.353	
G2	G2	-2.411		0.7920		0.2321	***	-3.352	***	-.1757E-04	
H2	H2	1.280		-.1840		-.2329		-.9431		1.602	
Y2	Y2	-.3433E-01		-1.751		-.3829		-.9769		1.230	
H5	H5	3.300		0.5174		-.6250		-.5392	***	0.8437	***
CD	CD	1.419		-1.558		-1.256	***	-.1824		-2.243	***
EB	EB	-.9636		0.9474		-1.270		1.105		-.5960	***
D4	D4	2.515		-1.038		-1.296		1.264		0.1114	
G1	G1	-.3419		4.910		-1.606	***	-.2975	***	0.1283	***
H6	H6	0.5299E-01		-.3520		-2.496		-.9560	***	1.956	
H3	H3	0.6778		-4.688		-2.803	***	0.3209	***	-.7302	***
SITE EFFECTS		-7.651	***	6.799	***	102.5	***	.		.	
AMMI1 SITE		0.1968		4.111		.		.		.	
AMMI2 SITE		0.4844E-01		-.5413		.		.		.	

FITTED VALUES FROM THE AMMI MODEL FILE 07_ELITE   17/ 8/ 9 21:27
---------- :PAGE 11

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
CC	CC	109.9	102.0	114.4	95.58	104.8	107.6	95.05
H1	H1	106.5	104.8	116.0	95.84	104.8	106.6	94.57
D3	D3	109.5	99.59	113.4	94.24	103.7	106.4	93.86
CE	CE	107.7	105.2	108.6	94.87	103.0	107.7	94.27
D1	D1	102.2	115.2	109.2	97.13	103.9	108.4	95.15
YC	YC	102.1	112.9	114.3	97.19	104.8	107.2	94.97
EA	EA	106.2	106.6	110.1	95.23	103.3	107.3	94.23
CA	CA	105.2	106.8	110.6	95.01	103.1	106.7	93.83
D2	D2	105.3	106.9	109.6	94.89	102.9	106.9	93.79
H4	H4	108.1	95.08	121.1	93.49	104.4	103.4	92.67
G2	G2	98.62	114.0	119.4	97.47	105.5	105.4	94.37
H2	H2	103.4	102.1	121.0	94.65	104.5	103.4	92.79
Y2	Y2	103.0	103.2	119.5	94.62	104.2	103.6	92.74
H5	H5	103.3	103.0	116.9	94.11	103.4	103.8	92.47
CD	CD	101.6	111.0	103.5	93.92	100.6	106.5	92.50
EB	EB	104.8	102.5	107.7	92.46	100.8	104.8	91.69
D4	D4	105.5	99.80	110.2	92.14	101.1	104.0	91.44
G1	G1	102.4	103.6	112.7	93.10	101.8	103.6	91.59
H6	H6	101.3	98.80	120.2	92.32	102.5	100.7	90.44
H3	H3	101.8	103.4	107.0	91.60	99.67	103.4	90.43
SITE ESTS.		104.4	104.8	113.3	94.49	103.1	105.4	93.14
AMMI1 SITE		1.800	-2.676	-1.757	-.8186	-.6313	0.7071E-01	-.2958
AMMI2 SITE		0.5692	-3.111	3.943	-.2370	0.6326	-1.054	-.2502

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	T-ESTS.	AMMI1 TRT	AMMI2 TRT
CC	CC	97.68	118.0	105.0	1.566	0.3564 I
H1	H1	96.52	111.4	104.1	0.1722	0.3787 I
D3	D3	96.69	118.2	103.9	1.876	0.5205 I
CE	CE	96.44	116.9	103.9	1.401	-.9013 I
D1	D1	95.71	106.4	103.7	-1.258	-1.891 I
YC	YC	95.65	104.0	103.7	-1.686	-.7806 I
EA	EA	96.07	113.5	103.6	0.6563	-.7770 I
CA	CA	95.52	111.6	103.1	0.3055	-.7012 I
D2	D2	95.51	112.3	103.1	0.4572	-.8604 I
H4	H4	95.64	112.9	103.0	1.065	2.353 I
G2	G2	94.46	95.80	102.8	-3.352	-.1757E-04 I
H2	H2	94.55	104.4	102.3	-.9431	1.602 I
Y2	Y2	94.38	104.3	102.2	-.9769	1.230 I
H5	H5	94.20	106.0	101.9	-.5392	0.8437 I
CD	CD	93.49	108.6	101.3	-.1824	-2.243 I
EB	EB	93.81	112.9	101.3	1.105	-.5960 I
D4	D4	93.85	113.2	101.2	1.264	0.1114 I
G1	G1	93.23	106.4	100.9	-.2975	0.1283 I
H6	H6	92.30	101.9	100.0	-.9560	1.956 I
H3	H3	92.12	108.3	99.74	0.3209	-.7302 I
SITE ESTS.		94.89	109.3	102.5	.	.
AMMI1 SITE		0.1968	4.111	.	.	.
AMMI2 SITE		0.4844E-01 I	-.5413 I	.	.	.

Addendum 11: Cross-site analysis of ethanol yield, the 2007 season trials

```
PBGXE - CROSS SITE ANALYSIS  FILE 07_ELITE   16/ 8/ 9 14:17
 ------------------------------------------------------------------ :PAGE   1

  20       CN$    CODES:
  1 CA          CA            2 CC          CC            3 CE          CE
  4 CD          CD            5 D1          D1            6 D2          D2
  7 D3          D3            8 D4          D4            9 YC          YC
 10 Y2          Y2           11 EA          EA           12 EB          EB
 13 G1          G1           14 G2          G2           15 H1          H1
 16 H2          H2           17 H3          H3           18 H4          H4
 19 H5          H5           20 H6          H6

   9     SITE$    CODES:
  1 PI 2007       2 KL 2007       3 LA 2007       4 ME 2007       5 RO 2007
  6 TY 2007       7 NA 2007       8 RI 2007       9 AL 2007

 TREATMENT BY VARIATE MEANS HAVE BEEN REQUESTED FOR  1 VARIATES:
 EYLH_ASI
 ROWS OF MEANS TABLES TO BE SORTED ON VARIATE EYLH_ASI
 GXE ANALYSIS HAS BEEN REQUESTED FOR  1 VARIATES
```

TREATMENT MEANS AND COUNTS OVER SITES FOR EACH VARIATE. FILE 07_ELITE 16/ 8/ 9 14:17
:PAGE 2

VARIETY\SITE		EYLH_ASI	
G2	G2	2787.	27
D2	D2	2736.	27
D1	D1	2703.	27
G1	G1	2691.	27
D4	D4	2678.	27
H1	H1	2669.	27
D3	D3	2655.	27
H3	H3	2637.	27
EA	EA	2632.	27
YC	YC	2621.	27
H2	H2	2604.	27
H6	H6	2590.	27
CC	CC	2587.	27
Y2	Y2	2545.	27
CA	CA	2510.	27
H4	H4	2472.	27
CE	CE	2457.	27
EB	EB	2186.	27
H5	H5	2137.	27
CD	CD	1817.	27
SITE MEANS		2536.	540

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
G2	G2	2782.	2088.	3473.	1738.	3921.	2917.	2616.
D2	D2	2549.	2512.	3019.	1794.	3451.	2790.	2592.
D1	D1	2558.	2564.	3148.	1957.	3418.	2928.	2512.
G1	G1	2444.	2115.	3445.	1478.	3830.	2581.	2564.
D4	D4	2459.	2411.	2880.	1513.	3756.	2572.	2699.
H1	H1	2705.	2071.	3462.	1568.	2878.	2702.	2856.
D3	D3	2611.	1979.	2930.	1556.	3609.	2735.	2711.
H3	H3	2518.	2216.	3002.	1817.	3102.	3166.	2484.
EA	EA	2382.	2330.	3411.	1910.	2862.	2722.	2463.
YC	YC	2550.	2534.	2913.	1328.	3511.	3030.	2333.
H2	H2	2651.	2317.	3178.	1436.	3031.	2626.	2833.
H6	H6	2529.	2045.	3125.	1415.	3322.	2899.	2852.
CC	CC	2302.	1956.	2877.	1605.	3384.	2725.	2580.
Y2	Y2	2412.	2070.	3401.	1791.	2709.	2838.	2795.
CA	CA	2511.	2277.	3143.	1455.	3631.	2770.	2629.
H4	H4	2596.	2251.	3212.	1237.	2633.	2662.	2134.
CE	CE	2549.	2341.	2466.	1684.	2949.	2432.	2139.
EB	EB	1877.	1987.	2682.	1446.	2554.	2310.	2014.
H5	H5	2380.	1928.	2674.	1370.	2978.	1819.	1638.
CD	CD	2603.	2832.	2290.	834.4	2716.	1825.	275.1
SITE MEANS		2498.	2241.	3037.	1547.	3212.	2652.	2386.
SITE INDEX		2498.	2241.	3037.	1547.	3212.	2652.	2386.

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	TRT MEANS
G2	G2	2395.	3150.	2787.
D2	D2	2658.	3254.	2736.
D1	D1	2197.	3042.	2703.
G1	G1	2317.	3442.	2691.
D4	D4	2456.	3352.	2678.
H1	H1	2376.	3406.	2669.
D3	D3	2482.	3284.	2655.
H3	H3	2341.	3090.	2637.
EA	EA	2289.	3323.	2632.
YC	YC	2374.	3016.	2621.
H2	H2	2354.	3007.	2604.
H6	H6	2339.	2789.	2590.
CC	CC	2377.	3477.	2587.
Y2	Y2	1954.	2931.	2545.
CA	CA	1515.	2661.	2510.
H4	H4	2521.	3006.	2472.
CE	CE	2533.	3019.	2457.
EB	EB	1991.	2817.	2186.
H5	H5	1920.	2529.	2137.
CD	CD	834.3	2143.	1817.
SITE MEANS		2211.	3037.	2536.
SITE INDEX		2211.	3037.	2536.

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

ENVIRONMENT	MEAN	SE	DUNCAN GROUPS	LSD TESTS
RO 2007	3212.2	66.233	|	.
AL 2007	3036.8	66.233	|	..
LA 2007	3036.5	66.233	|	...
TY 2007	2652.5	66.233	|	333.
PI 2007	2498.5	66.233	||	333..
NA 2007	2386.0	66.233	||	3332..
KL 2007	2241.2	66.233	|	33332..
RI 2007	2211.2	66.233	|	33332...
ME 2007	1546.6	66.233		33333333.

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

TREATMENT		MEAN	SE	DUNCAN GROUPS	LSD TESTS
G2	G2	2786.5	98.734	|	.
D2	D2	2735.6	98.734	|	..
D1	D1	2702.7	98.734	|	...
G1	G1	2690.7	98.734	|	....
D4	D4	2677.7	98.734	|	.....
H1	H1	2669.2	98.734	|	.....
D3	D3	2655.1	98.734	|	.....
H3	H3	2637.4	98.734	|	.....
EA	EA	2632.4	98.734	|	.....
YC	YC	2621.0	98.734	|	.....
H2	H2	2603.7	98.734	|	.....
H6	H6	2590.3	98.734	|	.....
CC	CC	2587.1	98.734	|	.....
Y2	Y2	2544.7	98.734	|	.....
CA	CA	2510.2	98.734	|	1.....
H4	H4	2472.4	98.734	||	1.....
CE	CE	2456.8	98.734	||	11.....
EB	EB	2186.5	98.734	||	3333333222222111..
H5	H5	2137.5	98.734	|	33333333332222211..
CD	CD	1817.0	98.734		3333333333333333321.

RESIDUALS FROM THE ADDITIVE TREATMENT BY SITE MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN STANDARD ERRORS, ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		R O	A L	L A	T Y	P I	N A	K L	R I	M E	T - E
		2	2	2	2	2	2	2	2	2	F
		0	0	0	0	0	0	0	0	0	C
		0	0	0	0	0	0	0	0	0	T
		7	7	7	7	7	7	7	7	7	S
G2	G2	1	0	0	0	0	0	-1	0	0	2
D2	D2	0	0	0	0	0	0	0	0	0	2
D1	D1	0	0	0	0	0	0	0	0	0	1
G1	G1	1	0	0	0	0	0	-1	0	0	1
D4	D4	1	0	-1	0	0	0	0	0	0	1
H1	H1	-1	0	1	0	0	1	-1	0	0	1
D3	D3	1	0	0	0	0	0	-1	0	0	1
H3	H3	0	0	0	1	0	0	0	0	0	1
EA	EA	-1	0	1	0	0	0	0	0	0	1
YC	YC	0	0	0	1	0	0	0	0	-1	0
H2	H2	0	0	0	0	0	1	0	0	0	0
H6	H6	0	-1	0	0	0	1	0	0	0	0
CC	CC	0	1	0	0	0	0	-1	0	0	0
Y2	Y2	-1	0	1	0	0	1	0	0	0	0
CA	CA	1	-1	0	0	0	0	0	-2	0	0
H4	H4	-1	0	0	0	0	0	0	1	0	0
CE	CE	0	0	-1	0	0	0	0	1	0	0
EB	EB	-1	0	0	0	-1	0	0	0	0	-3
H5	H5	0	0	0	-1	1	-1	0	0	0	-4
CD	CD	0	0	0	0	3	-5	4	-2	0	-7
SITE EFFECTS		10	0	8	1	0	-2	-4	-5	-15	114

```
BOX PLOT OF  180 STUDENTIZED RESIDUALS FROM LPLT= -2.464    TO ULPT=  3.026
NO.<LPLT                                                          NO.>UPLT
       1 --------------------I      +     I------------              *   1

MEDIAN=  0.2757E-01 ANDERSON-DARLING STATISTIC=  1.281 **

ANALYSIS OF VARIANCE FOR THE ADDITIVE MODEL

-------------------------------------------------------------------
SOURCE                  D.F.     S.S.         M.S.     F       FPROB
-------------------------------------------------------------------
TREATMENTS               19   0.947074E+07    498460.
LOCATIONS                 8   0.433501E+08   0.541876E+07
TREATMENT X SITES       152   0.133359E+08    87736.0
-------------------------------------------------------------------
TOTAL                   179   0.661567E+08
```

RESIDUALS FROM THE ADDITIVE SITE X TREATMENT MODEL FILE 07_ELITE   16/ 8/ 9 14:17
:PAGE   5

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
G2	G2	32.81	-404.2	185.4	-59.60	457.6	13.50	-20.68
D2	D2	-149.1	71.40	-217.4	47.53	39.39	-62.77	6.521
D1	D1	-107.1	155.5	-55.52	243.5	38.41	108.5	-40.52
G1	G1	-209.8	-281.1	253.3	-223.0	463.1	-226.9	23.44
D4	D4	-181.2	28.11	-298.3	-175.7	401.9	-222.0	171.5
H1	H1	72.91	-303.8	292.4	-112.2	-468.2	-84.41	336.3
D3	D3	-7.073	-381.5	-225.7	-110.1	277.3	-36.82	205.3
H3	H3	-81.67	-126.7	-136.3	169.1	-211.5	411.5	-3.988
EA	EA	-212.7	-8.015	278.0	266.5	-447.2	-27.35	-19.84
YC	YC	-34.07	207.3	-208.9	-303.4	214.0	292.4	-138.4
H2	H2	84.77	7.858	73.05	-178.5	-248.8	-94.23	379.0
H6	H6	-24.04	-251.1	33.95	-186.4	54.78	191.7	411.2
CC	CC	-247.5	-336.2	-210.6	6.755	120.3	21.56	142.6
Y2	Y2	-95.26	-180.0	355.4	235.3	-512.1	176.9	400.5
CA	CA	38.26	61.16	131.7	-65.72	444.6	143.0	268.3
H4	H4	160.9	72.97	239.0	-246.7	-516.0	72.82	-189.1
CE	CE	129.3	178.6	-491.4	215.9	-184.8	-141.2	-168.2
EB	EB	-272.3	95.10	-5.669	248.3	-308.5	7.151	-22.62
H5	H5	279.4	85.55	35.44	222.0	163.6	-434.9	-349.4
CD	CD	823.6 **	1309. ***	-27.76	6.565	222.1	-108.4	-1392. ***
SITE EFFECTS		-37.22	-294.5 ***	500.8 ***	-989.2 ***	676.5 ***	116.8	-149.8 *

SECTION 2

VARIETY\SITE		RI 2007		AL 2007		T-EFCTS	
G2	G2	-67.51		-137.3		250.8	*
D2	D2	247.2		17.23		199.9	*
D1	D1	-181.0		-161.7		167.0	
G1	G1	-48.97		250.1		154.9	
D4	D4	103.0		172.7		142.0	
H1	H1	31.38		235.5		133.5	
D3	D3	151.2		127.5		119.4	
H3	H3	27.82		-48.12		101.6	
EA	EA	-19.11		189.8		96.66	
YC	YC	77.29		-106.3		85.26	
H2	H2	74.85		-98.04		67.96	
H6	H6	72.79		-302.9		54.60	
CC	CC	114.7		388.4		51.36	
Y2	Y2	-266.1		-114.7		8.978	
CA	CA	-670.7	*	-350.7		-25.56	
H4	H4	373.5		32.60		-63.30	
CE	CE	401.0		60.88		-78.92	
EB	EB	129.3		129.3		-349.2	***
H5	H5	107.5		-109.2		-398.2	***
CD	CD	-658.1	*	-175.1		-718.7	***
SITE EFFECTS		-324.5	***	501.1		2536.	***

FITTED VALUES FROM ADDITIVE SITE X TREATMENT MODEL FILE 07_ELITE   16/ 8/ 9 14:17
 -- :PAGE   6

 SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
G2	G2	2749.	2492.	3287.	1797.	3463.	2903.	2637.
D2	D2	2698.	2441.	3236.	1746.	3412.	2852.	2586.
D1	D1	2665.	2408.	3203.	1714.	3379.	2819.	2553.
G1	G1	2653.	2396.	3191.	1701.	3367.	2807.	2541.
D4	D4	2641.	2383.	3179.	1689.	3354.	2795.	2528.
H1	H1	2632.	2375.	3170.	1680.	3346.	2786.	2519.
D3	D3	2618.	2361.	3156.	1666.	3332.	2772.	2505.
H3	H3	2600.	2343.	3138.	1648.	3314.	2754.	2488.
EA	EA	2595.	2338.	3133.	1643.	3309.	2749.	2483.
YC	YC	2584.	2326.	3122.	1632.	3297.	2738.	2471.
H2	H2	2566.	2309.	3104.	1615.	3280.	2720.	2454.
H6	H6	2553.	2296.	3091.	1601.	3267.	2707.	2441.
CC	CC	2550.	2293.	3088.	1598.	3264.	2704.	2437.
Y2	Y2	2507.	2250.	3046.	1556.	3221.	2661.	2395.
CA	CA	2473.	2216.	3011.	1521.	3187.	2627.	2360.
H4	H4	2435.	2178.	2973.	1483.	3149.	2589.	2323.
CE	CE	2420.	2162.	2958.	1468.	3133.	2574.	2307.
EB	EB	2149.	1892.	2687.	1197.	2863.	2303.	2037.
H5	H5	2100.	1843.	2638.	1148.	2814.	2254.	1988.
CD	CD	1780.	1522.	2318.	827.8	2493.	1934.	1667.
SITE ESTS.		2498.	2241.	3037.	1547.	3212.	2652.	2386.

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	T-ESTS.
G2	G2	2462.	3288.	2787.
D2	D2	2411.	3237.	2736.
D1	D1	2378.	3204.	2703.
G1	G1	2366.	3192.	2691.
D4	D4	2353.	3179.	2678.
H1	H1	2345.	3170.	2669.
D3	D3	2331.	3156.	2655.
H3	H3	2313.	3138.	2637.
EA	EA	2308.	3133.	2632.
YC	YC	2296.	3122.	2621.
H2	H2	2279.	3105.	2604.
H6	H6	2266.	3091.	2590.
CC	CC	2263.	3088.	2587.
Y2	Y2	2220.	3046.	2545.
CA	CA	2186.	3011.	2510.
H4	H4	2148.	2974.	2472.
CE	CE	2132.	2958.	2457.
EB	EB	1862.	2688.	2186.
H5	H5	1813.	2639.	2137.
CD	CD	1492.	2318.	1817.
SITE ESTS.		2211.	3037.	2536.

 REGRESSIONS OF EYLH_ASI FOR EACH VARIETY ON MEANS OF EYLH_ASI AT EACH SITE

VARIETY		MEAN	SLOPE	SE	MS-TXL	MS-REG	MS-DEV	R**2(%)
CA	CA	2510.16	1.199	0.230	111225.27	85598.68	114886.20	10.
CC	CC	2587.07	1.100	0.162	52253.48	21531.97	56642.26	5.
CE	CE	2456.80	0.662	0.149	72954.00	247880.67	47964.48	42.
CD	CD	1816.97	1.016	0.566	606978.56	574.62	693607.69	0.
D1	D1	2702.67	0.867	0.094	21473.50	38262.14	19075.12	22.
D2	D2	2735.61	0.896	0.089	17973.64	23274.82	17216.33	16.
D3	D3	2655.11	1.127	0.148	46022.30	35104.10	47582.04	10.
D4	D4	2677.74	1.137	0.161	54270.81	40498.52	56238.28	9.
YC	YC	2620.97	1.119	0.145	43440.86	30482.79	45292.01	9.
Y2	Y2	2544.69	0.836	0.215	95141.47	58168.01	100423.39	8.
EA	EA	2632.38	0.854	0.159	53893.33	45929.64	55031.00	11.
EB	EB	2186.47	0.793	0.109	34259.70	92582.84	25927.83	34.
G1	G1	2690.66	1.398*	0.122	71040.31	343223.72	32156.97	60.
G2	G2	2786.53	1.248	0.141	54465.97	132996.30	43247.36	31.
H1	H1	2669.23	1.035	0.197	73943.96	2603.32	84135.48	0.
H2	H2	2603.68	0.953	0.133	34265.68	4787.25	38476.88	2.
H3	H3	2637.35	0.851	0.126	35884.78	48052.09	34146.59	17.
H4	H4	2472.41	0.963	0.198	74635.48	3018.29	84866.50	1.
H5	H5	2137.47	0.901	0.177	62178.39	21287.14	68019.99	4.
H6	H6	2590.31	1.046	0.163	50682.64	4493.26	57281.13	1.

 SLOPE - SLOPES OF REGRESSIONS OF VARIETY MEANS ON SITE INDEX.
          * INDICATES SLOPES SIGNIFICANTLY DIFFERENT FROM THE
          SLOPE FOR THE OVERALL REGRESSION WHICH IS   1.00
 MS-TXL - CONTRIBUTION OF EACH VARIETY TO INTERACTION MS
 MS-REG - CONTRIBUTION OF EACH VARIETY TO THE REGRESSION
          COMPONENT OF THE TREATMENT BY LOCATION INTERACTION
 MS-DEV - DEVIATIONS FROM REGRESSION COMPONENT OF INTERACTION
 R**2   - SQUARED CORRELATION BETWEEN RESIDUALS FROM THE MAIN
          EFFECTS MODEL AND THE SITE INDEX.

```
VARIATE EYLH_ASI WAS SITE INDEX WITH OVERALL MEAN  2536.
THE FOLLOWING SITE MEANS OF EYLH_ASI WERE USED AS X-VARIATES
          2498.     2241.     3037.     1547.     3212.     2652.     2386.     2211.     3037.
-------------------------------------------------------------------
ANOVA FOR VARIABLE EYLH_ASI WITH SITE REGRESSIONS ON EYLH_ASI
-------------------------------------------------------------------
SOURCE             D.F.      S.S.         M.S.     F       FPROB
-------------------------------------------------------------------
TREATMENTS          19   0.947074E+07    498460.
LOCATIONS            8   0.433501E+08   0.541876E+07
TREATMENT X SITES  152   0.133359E+08    87736.0
   TRT X SITE REG   19   0.128035E+07    67386.9       0.743   0.769
   DEVIATIONS      133   0.120555E+08    90643.0
-------------------------------------------------------------------
TOTAL              179   0.661567E+08
```

```
AMMI ANALYSIS  FILE 07_ELITE   16/ 8/ 9 14:17
 ------------------------------------------------------------ :PAGE   8
 SINGULAR VALUES OF INTERACTION MATRIX (CONDITION=  0)
 2490.9    1578.4    1365.7    971.20    869.56    688.30    623.86    460.95

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR TREATMENTS

 CA CA          CA          -0.243416E+01-0.145240E+02-0.189157E+02 0.528644E+01
 CC CC          CC           0.854194E+01-0.419889E+01 0.850454E+01-0.448803E+00
 CE CE          CE          -0.357732E+01 0.712967E+01 0.161763E+02 0.707746E+01
 CD CD          CD          -0.439189E+02 0.133412E+01-0.383770E+01-0.280014E+01
 D1 D1          D1          -0.308883E+01-0.105469E+00-0.418597E+01 0.779151E+01
 D2 D2          D2           0.100880E+01-0.516670E-01 0.859419E+01 0.448430E+01
 D3 D3          D3           0.731669E+01-0.905493E+01 0.682489E+01-0.404545E+00
 D4 D4          D4           0.239059E+01-0.112814E+02 0.984105E+01 0.550080E+00
 YC YC          YC          -0.418387E+01-0.565916E+01 0.304206E+01 0.997203E+01
 Y2 Y2          Y2           0.838826E+01 0.110643E+02-0.163407E+02 0.124844E+01
 EA EA          EA           0.305092E+01 0.128666E+02-0.309696E+01-0.514084E+01
 EB EB          EB           0.208949E+01 0.962975E+01 0.341124E+01 0.258623E+01
 G1 G1          G1           0.439314E+01-0.125816E+02 0.113140E+01-0.146553E+02
 G2 G2          G2           0.275361E+01-0.124533E+02-0.396936E+01-0.615609E+01
 H1 H1          H1           0.981170E+01 0.951768E+01-0.461634E+01-0.107012E+02
 H2 H2          H2           0.509457E+01 0.409104E+01-0.338460E+01-0.722727E+00
 H3 H3          H3           0.293306E+01 0.529815E+01-0.142080E+01 0.115372E+02
 H4 H4          H4          -0.600261E+00 0.143446E+02 0.258269E+01-0.580123E+01
 H5 H5          H5          -0.837859E+01-0.906853E+00 0.567292E+01-0.986359E+01
 H6 H6          H6           0.840920E+01-0.445876E+01-0.601315E+01 0.616073E+01

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR ENVIRONMENTS

 PI             PI 2007     -0.174977E+02 0.216194E+01-0.300974E+01-0.565868E+01
 KL             KL 2007     -0.301192E+02 0.724585E+01 0.316349E+00 0.770161E+01
 LA             LA 2007      0.298944E+01 0.608364E+01-0.196242E+02-0.188500E+02
 ME             ME 2007     -0.166030E+01 0.969376E+01-0.337482E+00 0.490808E+01
 RO             RO 2007     -0.691578E+01-0.358274E+02 0.397339E+01-0.122419E+01
 TY             TY 2007      0.331055E+01 0.240218E+01-0.976163E+01 0.183399E+02
 NA             NA 2007      0.320957E+02-0.388710E+01-0.920592E+01 0.606602E+01
 RI             RI 2007      0.119463E+02 0.853963E+01 0.246009E+02-0.668669E-01
 AL             AL 2007      0.585094E+01 0.358751E+01 0.130484E+02-0.112159E+02
```

RESIDUALS FROM THE AMMI-2 MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN UNITS OF ROOT (RESIDUAL GXE MS), ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		ROW	ALLA	TYPIA	NALKL	RIME	T-EFCTS	
G2	G2	0	1	0	0	-1 0	0	EA
D2	D2	0	-1	0	0	0 1	0	EA
D1	D1	0	0	0	0	0 0	1	EA
G1	G1	0	1	0	0	0 0	0	EA
D4	D4	0	-1	0	0	0 0	0	EA
H1	H1	0	0	1	0	0 0	0	EA
D3	D3	0	0	0	0	0 0	0	EA
H3	H3	0	0	1	0	0 0	0	EA
EA	EA	0	0	0	0	0 0	0	EA
YC	YC	0	0	1	0	0 0	-1	EA
H2	H2	0	0	0	0	1 0	0	EA
H6	H6	0	-1	0	0	0 0	0	EA
CC	CC	0	1	0	0	0 0	0	EA
Y2	Y2	0	0	1	0	0 0	-2	EA
CA	CA	0	-1	1	0	0 1	-2	EA
H4	H4	0	0	0	0	0 0	1	-1 EA
CE	CE	0	0	-2	0	0 0	1	0 EA
EB	EB	0	0	0	0	-1 0	0	0 EA
H5	H5	0	0	0	-1	0 0	1	1 EA
CD	CD	0	0	0	0	0 0	0	0 EA

```
BOX PLOT OF  180 STANDERSIZED RESIDUALS FROM LPLT= -2.457    TO ULPT=  1.824
NO.<LPLT                                                          NO.>UPLT
       0 *   *   --------------------I     +       I-----------------   0
```

ANALYSIS OF VARIANCE FOR THE AMMI MODEL

SOURCE	D.F.	S.S.	M.S.	F	FPROB
TREATMENTS	19	0.947074E+07	498460.		
LOCATIONS	8	0.433501E+08	0.541876E+07		
TREATMENT X SITES	152	0.133359E+08	87736.0		
AMMI COMPONENT 1	26	0.620455E+07	238637.	4.216	0.000
AMMI COMPONENT 2	24	0.249146E+07	103811.	2.282	0.002
AMMI COMPONENT 3	22	0.186506E+07	84775.7	2.444	0.002
AMMI COMPONENT 4	20	943224.	47161.2	1.545	0.099
GXE RESIDUAL	60	0.183158E+07			
TOTAL	179	0.661567E+08			

 GENOTYPE MAP SHOWING BEST GENOTYPES OVER THE RANGE OF AMMI-2 SITE SCORES (2 CHRS/PIXEL)

```
     9.69 CDCDCDCDCDCDCDCDD1D1D1D1D1D1D1D1D1D1EAEAEAEAEAEAEAEAH1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
     8.83 CDCDCDCDCDCDCDCDD1D1D1D1D1D1D1D1D1D1D1D2D2D2EAEAEAH1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
     7.98 CDCDCDCDCDCDCDCDD1D1D1D1D1D1D1D1D1D1D1D2D2D2D2D2D2H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
     7.12 CDCDCDCDCDCDCDCDD1D1D1D1D1D1D1D1D1D1D1D2D2D2D2D2D2D2H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
     6.26 CDCDCDCDCDCDCDCDD1D1D1D1D1D1D1D1D1D1D1D2D2D2D2D2D2D2D2H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
     5.40 CDCDCDCDCDCDCDCDD1D1D1D1D1D1D1D1D1D1D1D2D2D2D2D2D2D2D2D2H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
     4.54 CDCDCDCDCDCDCDCDD1D1D1D1D1D1D1D1D1D1D1D2D2D2D2D2D2D2D2D2H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
     3.68 CDCDCDCDCDCDCDCDD1D1D1D1D1D1D1D1D1D1D1D2D2D2D2D2G2G2G2G2G2G2H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
     2.82 CDCDCDCDCDCDCDCDD1D1D1D1D1D1D1D1D1D1D1G2G2G2G2G2G2G2G2G2G2G2G2H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
     1.96 CDCDCDCDCDCDCDCDD1D1D1D1D1D1D1D1D1D1G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
     1.10 CDCDCDCDCDCDCDCDD1D1D1D1D1D1D1D1D1D1G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
     0.25 CDCDCDCDCDCDCDCDD1D1D1D1D1D1G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1H1
    -0.61 CDCDCDCDCDCDCDCDD1D1D1D1D1G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H1H1H1H1H1H1H1H1H1H1H1H1H1H1
    -1.47 CDCDCDCDCDCDCDCDD1D1D1G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H1H1H1H1H1H1H1H1H1H1H1H1H1
    -2.33 CDCDCDCDCDCDCDCDD1D1G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H1H1H1H1H1H1H1H1H1H1H1H1
    -3.19 CDCDCDCDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H1H1H1H1H1H1H1H1H1H1H1H1
    -4.05 CDCDCDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H1H1H1H1H1H1H1H1H1H1H1
    -4.91 CDCDCDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2H1H1H1H1H1H1H1H1H1
    -5.77 CDCDCDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    -6.63 CDCDCDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    -7.48 CDCDCDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    -8.34 CDCDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
    -9.20 CDCDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
   -10.06 CDCDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
   -10.92 CDCDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
   -11.78 CDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
   -12.64 CDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
   -13.50 CDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
   -14.36 CDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
   -15.21 CDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
   -16.07 CDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
   -16.93 CDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
   -17.79 CDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
   -18.65 CDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
   -19.51 CDCDCDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
   -20.37 CDCDCDG2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2G2
```

```
-21.23 CDCDCDG2G2G2G
```

AMMI RESIDUALS, ADDITIVE EFFECTS AND MULTIPLICATIVE SCORES FILE 07_ELITE 16/ 8/ 9 14:17
:PAGE 10

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
G2	G2	107.9	-231.0	252.9	65.70	30.43	34.30	-157.5
D2	D2	-131.3	102.2	-220.1	49.71	44.52	-65.99	-26.06
D1	D1	-160.9	63.21	-45.64	239.4	13.27	119.0	58.20
G1	G1	-105.8	-57.62	316.7	-93.78	42.67	-211.2	-166.5
D4	D4	-115.0	181.9	-236.8	-62.35	14.28	-202.9	50.93
H1	H1	224.0	-77.20	205.2	-188.2	-59.30	-139.8	58.43
D3	D3	140.5	-95.54	-192.5	-10.17	3.463	-39.29	-64.71
H3	H3	-41.81	-76.78	-177.3	122.6	-1.443	389.1	-77.53
EA	EA	-187.1	-9.353	190.6	146.8	34.83	-68.36	-67.75
YC	YC	-95.05	122.3	-161.9	-255.5	-17.64	319.9	-26.16
H2	H2	165.1	131.7	32.94	-209.7	-67.03	-120.9	231.4
H6	H6	132.7	34.49	35.94	-129.2	-46.81	174.6	124.0
CC	CC	-88.95	-48.54	-210.6	61.64	28.95	3.369	-147.8
Y2	Y2	27.59	-7.566	263.0	142.0	-57.68	122.5	174.3
CA	CA	27.06	93.08	227.3	71.03	-92.54	186.0	290.0
H4	H4	119.4	-49.05	153.5	-386.8	-6.211	40.35	-114.1
CE	CE	51.24	19.23	-524.1 *	140.8	45.93	-146.5	-25.66
EB	EB	-256.6	88.26	-70.50	158.4	50.93	-22.90	-52.25
H5	H5	134.8	-160.2	66.00	216.9	73.20	-405.0	-84.04
CD	CD	52.26	-23.33	95.41	-79.29	-33.82	33.80	22.70
SITE EFFECTS		-37.22 ***	-294.5	500.8	-989.2	676.5	116.8	-149.8 ***
AMMI1 SITE		-17.50 ***	-30.12 ***	2.989	-1.660 ***	-6.916 ***	3.311	32.10
AMMI2 SITE		2.162 ***	7.246	6.084	9.694 ***	-35.83	2.402	-3.887

SECTION 2

VARIETY\SITE		RI 2007		AL 2007		T-EFCTS		AMMI1 TRT		AMMI2 TRT	
G2	G2	5.942		-108.8		250.8	***	2.754	***	-12.45	
D2	D2	235.6		11.51		199.9		1.009	***	-.5167E-01	
D1	D1	-143.2		-143.2		167.0	***	-3.089		-.1055	
G1	G1	5.987		269.5		154.9	***	4.393		-12.58	
D4	D4	170.7		199.2		142.0		2.391	***	-11.28	***
H1	H1	-167.1		143.9		133.5	***	9.812		9.518	
D3	D3	141.1		117.1		119.4	***	7.317		-9.055	
H3	H3	-52.46		-84.29		101.6	***	2.933	***	5.298	***
EA	EA	-165.4		125.8		96.66	***	3.051		12.87	
YC	YC	175.6		-61.49		85.26	***	-4.184		-5.659	
H2	H2	-20.95		-142.5		67.96	***	5.095	***	4.091	***
H6	H6	10.40		-336.1		54.60	***	8.409	***	-4.459	***
CC	CC	48.52		353.5		51.36	***	8.542	***	-4.199	
Y2	Y2	-460.8	*	-203.4		8.978	***	8.388		11.06	***
CA	CA	-517.5	*	-284.4		-25.56	***	-2.434	***	-14.52	
H4	H4	258.2		-15.35		-63.30		-.6003	***	14.34	***
CE	CE	382.9		56.23		-78.92	***	-3.577		7.130	
EB	EB	22.15		82.52		-349.2		2.089	***	9.630	***
H5	H5	215.3		-56.88		-398.2		-8.379		-.9069	***
CD	CD	-144.9		77.13		-718.7		-43.92		1.334	
SITE EFFECTS		-324.5		501.1		2536.	***	.		.	
AMMI1 SITE		11.95		5.851	***	.		.		.	
AMMI2 SITE		8.540		3.588	***	.		.		.	

FITTED VALUES FROM THE AMMI MODEL FILE 07_ELITE   16/ 8/ 9 14:17
---------- :PAGE 11

SECTION 1

VARIETY\SITE		PI 2007	KL 2007	LA 2007	ME 2007	RO 2007	TY 2007	NA 2007
G2	G2	2674.	2319.	3220.	1672.	3890.	2883.	2774.
D2	D2	2681.	2410.	3239.	1744.	3407.	2856.	2618.
D1	D1	2719.	2500.	3194.	1718.	3404.	2809.	2454.
G1	G1	2549.	2173.	3128.	1572.	3788.	2792.	2731.
D4	D4	2574.	2229.	3117.	1575.	3742.	2775.	2649.
H1	H1	2481.	2148.	3257.	1756.	2937.	2841.	2797.
D3	D3	2470.	2075.	3123.	1566.	3605.	2774.	2775.
H3	H3	2560.	2293.	3179.	1695.	3104.	2777.	2561.
EA	EA	2570.	2339.	3221.	1763.	2827.	2790.	2531.
YC	YC	2645.	2411.	3075.	1584.	3529.	2710.	2359.
H2	H2	2486.	2185.	3145.	1646.	3098.	2747.	2602.
H6	H6	2396.	2010.	3089.	1544.	3368.	2724.	2728.
CC	CC	2391.	2005.	3088.	1543.	3355.	2722.	2728.
Y2	Y2	2385.	2078.	3138.	1649.	2767.	2716.	2621.
CA	CA	2484.	2184.	2915.	1384.	3724.	2584.	2339.
H4	H4	2477.	2300.	3059.	1623.	2639.	2622.	2248.
CE	CE	2498.	2322.	2990.	1543.	2903.	2579.	2165.
EB	EB	2134.	1899.	2752.	1287.	2503.	2333.	2066.
H5	H5	2245.	2089.	2608.	1153.	2904.	2224.	1722.
CD	CD	2551.	2855.	2195.	913.7	2749.	1792.	252.4
SITE ESTS.		2498.	2241.	3037.	1547.	3212.	2652.	2386.
AMMI1 SITE		-17.50	-30.12	2.989	-1.660	-6.916	3.311	32.10
AMMI2 SITE		2.162	7.246	6.084	9.694	-35.83	2.402	-3.887

SECTION 2

VARIETY\SITE		RI 2007	AL 2007	T-ESTS.	AMMI1 TRT	AMMI2 TRT
G2	G2	2389.	3259.	2787.	2.754	-12.45 I
D2	D2	2423.	3242.	2736.	1.009	-.5167E-01 I
D1	D1	2340.	3185.	2703.	-3.089	-.1055 I
G1	G1	2311.	3172.	2691.	4.393	-12.58 I
D4	D4	2285.	3152.	2678.	2.391	-11.28 I
H1	H1	2543.	3262.	2669.	9.812	9.518 I
D3	D3	2341.	3167.	2655.	7.317	-9.055 I
H3	H3	2393.	3175.	2637.	2.933	5.298 I
EA	EA	2454.	3198.	2632.	3.051	12.87 I
YC	YC	2198.	3077.	2621.	-4.184	-5.659 I
H2	H2	2375.	3149.	2604.	5.095	4.091 I
H6	H6	2328.	3125.	2590.	8.409	-4.459 I
CC	CC	2329.	3123.	2587.	8.542	-4.199 I
Y2	Y2	2415.	3135.	2545.	8.388	11.06 I
CA	CA	2033.	2945.	2510.	-2.434	-14.52 I
H4	H4	2263.	3021.	2472.	-.6003	14.34 I
CE	CE	2150.	2963.	2457.	-3.577	7.130 I
EB	EB	1969.	2734.	2186.	2.089	9.630 I
H5	H5	1705.	2586.	2137.	-8.379	-.9069 I
CD	CD	979.2	2066.	1817.	-43.92	1.334 I
SITE ESTS.		2211.	3037.	2536.	.	.
AMMI1 SITE		11.95	5.851	.	.	.
AMMI2 SITE		8.540 I	3.588 I	.	.	.

Addendum 12: Cross-site analysis of test weight, the 2007 season trials

```
PBGXE - CROSS SITE ANALYSIS  FILE 07_TSTW   16/ 7/ 9 16:37
 ------------------------------------------------------------------ :PAGE   1

  20       CN$    CODES:
  1 CA          CA            2 CC          CC            3 CE          CE
  4 CD          CD            5 D1          D1            6 D2          D2
  7 D3          D3            8 D4          D4            9 YC          YC
 10 Y2          Y2           11 EA          EA           12 EB          EB
 13 G1          G1           14 G2          G2           15 H1          H1
 16 H2          H2           17 H3          H3           18 H4          H4
 19 H5          H5           20 H6          H6

   9      SET$    CODES:
  1 AL           2 KL           3 LA           4 ME           5 NA
  6 PI           7 RI           8 RO           9 TY

 TREATMENT BY VARIATE MEANS HAVE BEEN REQUESTED FOR  1 VARIATES:
 TST_WGHT
 ROWS OF MEANS TABLES TO BE SORTED ON VARIATE TST_WGHT
 GXE ANALYSIS HAS BEEN REQUESTED FOR  1 VARIATES
```

TREATMENT MEANS AND COUNTS OVER SITES FOR EACH VARIATE. FILE 07_TSTW 16/ 7/ 9 16:37

:PAGE 2

VARIETY\SITE		TST_WGHT	
EB	EB	75.85	9
Y2	Y2	75.70	9
EA	EA	74.67	9
H4	H4	74.59	9
G1	G1	74.44	9
H3	H3	74.22	9
G2	G2	73.93	9
H6	H6	73.85	9
H5	H5	73.48	9
H1	H1	73.41	9
YC	YC	73.33	9
D3	D3	73.26	9
CA	CA	73.11	9
H2	H2	72.89	9
D2	D2	72.52	9
D1	D1	72.44	9
CD	CD	72.38	9
CE	CE	72.15	9
D4	D4	71.78	9
CC	CC	71.19	9
SITE MEANS		73.46	180

SECTION 1

VARIETY\SITE		AL	KL	LA	ME	NA	PI	RI
EB	EB	75.33	72.67	76.00	74.67	78.00	78.00	74.00
Y2	Y2	74.00	72.67	76.00	75.33	78.00	77.33	74.67
EA	EA	74.00	72.00	74.00	74.67	76.00	76.00	74.67
H4	H4	74.00	72.67	74.00	72.00	76.00	76.67	74.67
G1	G1	73.33	74.00	74.00	72.00	75.33	76.00	73.33
H3	H3	72.67	72.00	74.00	72.00	76.00	76.67	74.67
G2	G2	73.33	72.00	74.00	71.33	75.33	75.33	72.67
H6	H6	73.33	72.67	74.00	71.33	76.00	73.33	74.67
H5	H5	72.00	72.00	72.00	72.00	74.67	76.00	74.00
H1	H1	72.67	70.67	74.00	71.33	76.00	73.33	74.00
YC	YC	72.67	72.00	74.00	70.00	76.67	76.00	72.67
D3	D3	73.33	72.67	74.00	70.00	76.00	74.00	72.00
CA	CA	70.00	70.67	74.00	73.33	73.33	76.00	70.00
H2	H2	73.33	70.00	72.67	70.00	74.67	74.00	72.67
D2	D2	72.00	70.67	72.67	70.00	74.67	72.67	72.67
D1	D1	71.33	70.67	72.00	70.00	75.33	74.00	71.33
CD	CD	72.00	74.67	74.67	69.33	65.79	78.00	68.95
CE	CE	71.33	69.33	71.33	71.33	74.00	74.00	72.00
D4	D4	71.33	70.00	72.00	70.00	74.00	72.00	71.33
CC	CC	70.00	68.67	70.67	70.67	72.00	74.00	70.00
SITE MEANS		72.60	71.63	73.50	71.57	74.89	75.17	72.75
SITE INDEX		72.60	71.63	73.50	71.57	74.89	75.17	72.75
SE OF MEANS		0.6269	0.4726	0.3208	0.5814	0.4265	0.5309	0.7946
LSD(5%)		1.795	1.353	0.9184	1.664	1.223	1.520	2.277

SECTION 2

VARIETY\SITE		RO	TY	TRT MEANS
EB	EB	76.00	78.00	75.85
Y2	Y2	76.00	77.33	75.70
EA	EA	74.67	76.00	74.67
H4	H4	76.00	75.33	74.59
G1	G1	75.33	76.67	74.44
H3	H3	74.00	76.00	74.22
G2	G2	76.67	74.67	73.93
H6	H6	72.67	76.67	73.85
H5	H5	74.00	74.67	73.48
H1	H1	74.00	74.67	73.41
YC	YC	70.67	75.33	73.33
D3	D3	72.00	75.33	73.26
CA	CA	76.00	74.67	73.11
H2	H2	74.00	74.67	72.89
D2	D2	73.33	74.00	72.52
D1	D1	73.33	74.00	72.44
CD	CD	76.67	71.33	72.38
CE	CE	72.00	74.00	72.15
D4	D4	71.33	74.00	71.78
CC	CC	72.00	72.67	71.19
SITE MEANS		74.03	75.00	73.46
SITE INDEX		74.03	75.00	73.46
SE OF MEANS		0.7940	0.4537	.
LSD(5%)		2.273	1.299	.

ANALYSIS OF RESIDUAL VARIATION WITHIN SITES
POOLED ERROR MEAN SQUARES FOR 9 SITES WITH 339 D.F.= 0.99626
BARTLETT'S STATISTIC=   49.95 P-VALUE (CHI^2 WITH 8 D.F.) = 1.000

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

ENVIRONMENT	MEAN	SE	DUNCAN GROUPS	LSD TESTS
PI	75.167	0.29784	|	.
TY	75.000	0.29784	|	..
NA	74.889	0.29784	|	...
RO	74.033	0.29784	|	211.
LA	73.500	0.29784	||	332..
RI	72.747	0.29784	||	3332..
AL	72.600	0.29784	|	33331..
KL	71.633	0.29784	|	3333321.
ME	71.567	0.29784	|	3333321..

 PREDICTED MEANS, SES AND MULTIPLE COMPARISONS

TREATMENT		MEAN	SE	DUNCAN GROUPS	LSD TESTS
EB	EB	75.852	0.44399	|	.
Y2	Y2	75.704	0.44399	||	..
EA	EA	74.667	0.44399	|||	...
H4	H4	74.593	0.44399	|||	1...
G1	G1	74.444	0.44399	|||	11...
H3	H3	74.222	0.44399	|||	11....
G2	G2	73.926	0.44399	||||	22.....
H6	H6	73.852	0.44399	|||||	22.....
H5	H5	73.481	0.44399	|||||	33.....
H1	H1	73.407	0.44399	|||||	331.....
YC	YC	73.333	0.44399	|||||	3311.....
D3	D3	73.259	0.44399	|||||	3311.....
CA	CA	73.111	0.44399	|||||	33111.....
H2	H2	72.889	0.44399	|||||	332211.....
D2	D2	72.519	0.44399	|||||	33322211.....
D1	D1	72.444	0.44399	||||	33332211.....
CD	CD	72.378	0.44399	|||	33332211.....
CE	CE	72.148	0.44399	|||	3333322211.....
D4	D4	71.778	0.44399	||	3333333221111.....
CC	CC	71.185	0.44399	|	3333333333322211....

RESIDUALS FROM THE ADDITIVE TREATMENT BY SITE MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN STANDARD ERRORS, ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		P I	T Y	N A	R O	L A	R I	A L	K L	M E	T - E F C T S
EB	EB	0	0	0	0	0	0	0	-1	0	5
Y2	Y2	0	0	0	0	0	0	0	0	1	5
EA	EA	0	0	0	0	0	0	0	0	1	2
H4	H4	0	0	0	0	0	0	0	0	0	2
G1	G1	0	0	0	0	0	0	0	1	0	2
H3	H3	0	0	0	0	0	0	0	0	0	1
G2	G2	0	0	0	1	0	0	0	0	0	1
H6	H6	-1	1	0	-1	0	1	0	0	0	1
H5	H5	0	0	0	0	-1	1	0	0	0	0
H1	H1	-1	0	0	0	0	1	0	0	0	0
YC	YC	0	0	1	-2	0	0	0	0	-1	0
D3	D3	0	0	1	-1	0	0	0	1	-1	0
CA	CA	0	0	0	1	0	-1	-1	0	1	0
H2	H2	0	0	0	0	0	0	1	0	0	-1
D2	D2	-1	0	0	0	0	0	0	0	0	-2
D1	D1	0	0	1	0	0	0	0	0	0	-2
CD	CD	3	-2	-6	3	1	-2	0	3	0	-2
CE	CE	0	0	0	0	0	0	0	0	0	-3
D4	D4	-1	0	0	0	0	0	0	0	0	-3
CC	CC	0	0	0	0	0	0	0	0	1	-5
SITE EFFECTS		6	0	5	2	0	-2	-3	-6	-6	739

```
BOX PLOT OF  180 STUDENTIZED RESIDUALS FROM LPLT= -2.647    TO ULPT=  3.362
NO.<LPLT                                                          NO.>UPLT
       1 *   ** -------------I    +    I-------------          ** *   0

MEDIAN=  0.3209E-01 ANDERSON-DARLING STATISTIC=  3.180 **
```

ANALYSIS OF VARIANCE FOR THE ADDITIVE MODEL

SOURCE	D.F.	S.S.	M.S.	F	FPROB
TREATMENTS	19	258.623	13.6117		
LOCATIONS	8	316.538	39.5672		
TREATMENT X SITES	152	269.669	1.77414		
POOLED ERROR(PER MEAN)	339	112.578	0.332088		
TOTAL	179	844.830			

ESTIMATED MAXIMUM STRUCTURAL CONTENT OF TREATMENT X SITE SS IS 81.28%

RESIDUALS FROM THE ADDITIVE SITE X TREATMENT MODEL FILE 07_TSTW 16/ 7/ 9 16:37
:PAGE 5

SECTION 1

VARIETY\SITE		AL	KL	LA	ME	NA	PI	RI
EB	EB	0.3411	-1.359	0.1078	0.7078	0.7183	0.4411	-1.140
Y2	Y2	-.8441	-1.211	0.2559	1.523	0.8665	-.7739E-01	-.3248
EA	EA	0.1930	-.8403	-.7070	1.893	-.9649E-01	-.3737	0.7123
H4	H4	0.2671	-.9961E-01	-.6329	-.6996	-.2242E-01	0.3671	0.7864
G1	G1	-.2515	1.382	-.4848	-.5515	-.5409	-.1515	-.3988
H3	H3	-.6959	-.3959	-.2626	-.3292	0.3480	0.7374	1.157
G2	G2	0.2671	-.9961E-01	0.3372E-01	-.6996	-.2241E-01	-.2996	-.5470
H6	H6	0.3411	0.6411	0.1078	-.6255	0.7183	-2.226	1.527
H5	H5	-.6218	0.3448	-1.522	0.4115	-.2446	0.8115	1.231
H1	H1	0.1189	-.9144	0.5522	-.1811	1.163	-1.781	1.305
YC	YC	0.1930	0.4930	0.6263	-1.440	1.904	0.9597	0.4561E-01
D3	D3	0.9337	1.234	0.7004	-1.366	1.311	-.9663	-.5470
CA	CA	-2.251	-.6181	0.8485	2.115	-1.208	1.182	-2.399
H2	H2	1.304	-1.063	-.2626	-.9959	0.3480	-.5959	0.4901
D2	D2	0.3411	-.2554E-01	0.1078	-.6255	0.7183	-1.559	0.8604
D1	D1	-.2515	0.4854E-01	-.4848	-.5515	1.459	-.1515	-.3988
CD	CD	0.4815	4.115 ***	2.248	-1.152	-8.019 ***	3.915 **	-2.719 *
CE	CE	0.4483E-01	-.9885	-.8552	1.078	0.4220	0.1448	0.5641
D4	D4	0.4152	0.4854E-01	0.1819	0.1152	0.7924	-1.485	0.2678
CC	CC	-.3255	-.6922	-.5589	1.374	-.6150	1.108	-.4729
SITE EFFECTS		-.8597 **	-1.826 ***	0.4035E-01	-1.893 ***	1.430 ***	1.707 ***	-.7123 *

SECTION 2

VARIETY\SITE		RO		TY		T-EFCTS	
EB	EB	-.4255		0.6078		2.392	***
Y2	Y2	-.2774		0.8928E-01		2.244	***
EA	EA	-.5737		-.2070		1.207	**
H4	H4	0.8337		-.7996		1.133	**
G1	G1	0.3152		0.6819		0.9848	*
H3	H3	-.7959		0.2374		0.7626	
G2	G2	2.167		-.7996		0.4663	
H6	H6	-1.759		1.274		0.3922	
H5	H5	-.5517E-01		-.3552		0.2183E-01	
H1	H1	0.1891E-01		-.2811		-.5224E-01	
YC	YC	-3.240	**	0.4597		-.1263	
D3	D3	-1.833		0.5337		-.2004	
CA	CA	2.315		0.1520E-01		-.3485	
H2	H2	0.5374		0.2374		-.5708	
D2	D2	0.2411		-.5887E-01		-.9411	*
D1	D1	0.3152		0.1520E-01		-1.015	*
CD	CD	3.715	**	-2.585	*	-1.081	*
CE	CE	-.7218		0.3115		-1.312	**
D4	D4	-1.018		0.6819		-1.682	***
CC	CC	0.2411		-.5887E-01		-2.274	***
SITE EFFECTS		0.5737	*	1.540		73.46	***

FITTED VALUES FROM ADDITIVE SITE X TREATMENT MODEL FILE 07_TSTW 16/ 7/ 9 16:37
 ---------- :PAGE 6

 SECTION 1

VARIETY\SITE		AL	KL	LA	ME	NA	PI	RI
EB	EB	74.99	74.03	75.89	73.96	77.28	77.56	75.14
Y2	Y2	74.84	73.88	75.74	73.81	77.13	77.41	74.99
EA	EA	73.81	72.84	74.71	72.77	76.10	76.37	73.95
H4	H4	73.73	72.77	74.63	72.70	76.02	76.30	73.88
G1	G1	73.58	72.62	74.48	72.55	75.87	76.15	73.73
H3	H3	73.36	72.40	74.26	72.33	75.65	75.93	73.51
G2	G2	73.07	72.10	73.97	72.03	75.36	75.63	73.21
H6	H6	72.99	72.03	73.89	71.96	75.28	75.56	73.14
H5	H5	72.62	71.66	73.52	71.59	74.91	75.19	72.77
H1	H1	72.55	71.58	73.45	71.51	74.84	75.11	72.70
YC	YC	72.47	71.51	73.37	71.44	74.76	75.04	72.62
D3	D3	72.40	71.43	73.30	71.37	74.69	74.97	72.55
CA	CA	72.25	71.28	73.15	71.22	74.54	74.82	72.40
H2	H2	72.03	71.06	72.93	71.00	74.32	74.60	72.18
D2	D2	71.66	70.69	72.56	70.63	73.95	74.23	71.81
D1	D1	71.58	70.62	72.48	70.55	73.87	74.15	71.73
CD	CD	71.52	70.55	72.42	70.49	73.81	74.09	71.67
CE	CE	71.29	70.32	72.19	70.26	73.58	73.86	71.44
D4	D4	70.92	69.95	71.82	69.88	73.21	73.48	71.07
CC	CC	70.33	69.36	71.23	69.29	72.62	72.89	70.47
SITE ESTS.		72.60	71.63	73.50	71.57	74.89	75.17	72.75

SECTION 2

VARIETY\SITE		RO	TY	T-ESTS.
EB	EB	76.43	77.39	75.85
Y2	Y2	76.28	77.24	75.70
EA	EA	75.24	76.21	74.67
H4	H4	75.17	76.13	74.59
G1	G1	75.02	75.98	74.44
H3	H3	74.80	75.76	74.22
G2	G2	74.50	75.47	73.93
H6	H6	74.43	75.39	73.85
H5	H5	74.06	75.02	73.48
H1	H1	73.98	74.95	73.41
YC	YC	73.91	74.87	73.33
D3	D3	73.83	74.80	73.26
CA	CA	73.68	74.65	73.11
H2	H2	73.46	74.43	72.89
D2	D2	73.09	74.06	72.52
D1	D1	73.02	73.98	72.44
CD	CD	72.95	73.92	72.38
CE	CE	72.72	73.69	72.15
D4	D4	72.35	73.32	71.78
CC	CC	71.76	72.73	71.19
SITE ESTS.		74.03	75.00	73.46

 REGRESSIONS OF TST_WGHT FOR EACH VARIETY ON MEANS OF TST_WGHT AT EACH SITE

VARIETY		MEAN	SLOPE	SE	MS-TXL	MS-REG	MS-DEV	R**2(%)
CA	CA	73.11	1.155	0.468	3.08	0.38	3.46	2.
CC	CC	71.19	1.020	0.205	0.58	0.01	0.67	0.
CE	CE	72.15	1.013	0.190	0.50	0.00	0.57	0.
CD	CD	72.38	0.346	1.059	16.38	6.77	17.75	5.
D1	D1	72.44	1.219	0.143	0.38	0.76	0.32	25.
D2	D2	72.52	0.921	0.194	0.53	0.10	0.60	2.
D3	D3	73.26	0.996	0.320	1.42	0.00	1.62	0.
D4	D4	71.78	0.887	0.200	0.58	0.20	0.63	4.
YC	YC	73.33	1.307	0.386	2.25	1.49	2.36	8.
Y2	Y2	75.70	1.087	0.221	0.69	0.12	0.77	2.
EA	EA	74.67	0.736	0.207	0.73	1.10	0.68	19.
EB	EB	75.85	1.261	0.189	0.63	1.08	0.56	21.
G1	G1	74.44	0.949	0.178	0.44	0.04	0.50	1.
G2	G2	73.93	1.072	0.237	0.79	0.08	0.89	1.
H1	H1	73.41	0.950	0.259	0.94	0.04	1.06	1.
H2	H2	72.89	1.158	0.203	0.62	0.39	0.65	8.
H3	H3	74.22	1.175	0.165	0.44	0.49	0.43	14.
H4	H4	74.59	1.034	0.166	0.38	0.02	0.43	1.
H5	H5	73.48	0.914	0.217	0.67	0.12	0.75	2.
H6	H6	73.85	0.799	0.340	1.68	0.64	1.83	5.

 SLOPE - SLOPES OF REGRESSIONS OF VARIETY MEANS ON SITE INDEX.
          * INDICATES SLOPES SIGNIFICANTLY DIFFERENT FROM THE
          SLOPE FOR THE OVERALL REGRESSION WHICH IS   1.00
 MS-TXL - CONTRIBUTION OF EACH VARIETY TO INTERACTION MS
 MS-REG - CONTRIBUTION OF EACH VARIETY TO THE REGRESSION
          COMPONENT OF THE TREATMENT BY LOCATION INTERACTION
 MS-DEV - DEVIATIONS FROM REGRESSION COMPONENT OF INTERACTION
 R**2   - SQUARED CORRELATION BETWEEN RESIDUALS FROM THE MAIN
          EFFECTS MODEL AND THE SITE INDEX.

```
VARIATE TST_WGHT WAS SITE INDEX WITH OVERALL MEAN  73.46
THE FOLLOWING SITE MEANS OF TST_WGHT WERE USED AS X-VARIATES
          72.60     71.63     73.50     71.57     74.89     75.17     72.75     74.03     75.00
-----------------------------------------------------------------
ANOVA FOR VARIABLE TST_WGHT WITH SITE REGRESSIONS ON TST_WGHT
-----------------------------------------------------------------
SOURCE             D.F.      S.S.         M.S.     F       FPROB
-------------------------------------------------------------------
TREATMENTS          19    258.623        13.6117
LOCATIONS            8    316.538        39.5672
TREATMENT X SITES  152    269.669        1.77414
   TRT X SITE REG   19    13.8354       0.728177       0.379   0.991
   DEVIATIONS      133    255.834        1.92356
-----------------------------------------------------------------
TOTAL              179    844.830
```

```
AMMI ANALYSIS  FILE 07_TSTW   16/ 7/ 9 16:37
 ------------------------------------------------------------ :PAGE   8
 SINGULAR VALUES OF INTERACTION MATRIX (CONDITION=  0)
 12.831    6.6266    4.6549    3.9533    3.2428    2.6191    2.0791    1.4549

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR TREATMENTS

 CA CA          CA          -0.804142E+00-0.136228E+01-0.313495E+00 0.796345E+00
 CC CC          CC          -0.216281E+00-0.669794E+00-0.315120E+00-0.315744E+00
 CE CE          CE           0.333477E+00-0.423846E+00-0.169273E+00-0.490963E+00
 CD CD          CD          -0.314362E+01 0.749817E+00-0.941949E-01-0.136220E+00
 D1 D1          D1           0.232991E+00-0.108656E-01 0.698492E-01 0.359358E+00
 D2 D2          D2           0.329259E+00 0.289138E+00 0.666050E+00 0.160007E+00
 D3 D3          D3           0.423391E+00 0.977215E+00-0.426086E+00 0.620697E+00
 D4 D4          D4           0.477361E+00 0.245838E+00 0.415104E-01 0.227464E+00
 YC YC          YC           0.598776E+00 0.829229E+00-0.127629E+01 0.430294E-01
 Y2 Y2          Y2           0.269234E+00-0.737040E+00-0.280790E+00 0.205600E+00
 EA EA          EA           0.239360E+00-0.538497E+00 0.286462E-01-0.594271E+00
 EB EB          EB           0.196997E+00-0.467188E+00-0.449348E+00 0.415662E+00
 G1 G1          G1          -0.207399E+00 0.323266E+00-0.673420E-02 0.711860E-01
 G2 G2          G2          -0.314943E+00-0.896450E-01 0.801011E+00 0.489940E+00
 H1 H1          H1           0.535924E+00 0.445468E-01 0.706545E+00 0.158732E+00
 H2 H2          H2           0.219566E+00 0.173113E+00 0.633726E+00 0.740914E-01
 H3 H3          H3           0.223907E+00 0.109618E-01-0.272940E+00-0.559352E+00
 H4 H4          H4          -0.937602E-01 0.753980E-01 0.479130E+00-0.392592E+00
 H5 H5          H5          -0.148119E-01-0.169672E+00 0.191141E-02-0.102114E+01
 H6 H6          H6           0.714710E+00 0.750309E+00 0.175893E+00-0.111826E+00

 SCORES FOR FIRST 4 AMMI COMPONENTS FOR ENVIRONMENTS

 AL             AL           0.110781E+00 0.869486E+00 0.414665E+00-0.424910E-01
 KL             KL          -0.102671E+01 0.136569E+01-0.275654E+00-0.479202E-01
 LA             LA          -0.536473E+00 0.422707E+00-0.281696E+00 0.965523E+00
 ME             ME           0.640712E-01-0.168453E+01-0.335530E+00-0.302970E+00
 NA             NA           0.240572E+01-0.194463E+00-0.137368E+00 0.635237E+00
 PI             PI          -0.132449E+01-0.356871E+00-0.118457E+01-0.618307E+00
 RI             RI           0.101768E+01 0.390791E+00 0.761686E+00-0.135328E+01
 RO             RO          -0.150491E+01-0.819246E+00 0.143399E+01 0.471495E+00
 TY             TY           0.794332E+00 0.643857E-02-0.395525E+00 0.292708E+00
```

RESIDUALS FROM THE AMMI-2 MODEL

(ENTRIES ARE SIZE OF RESIDUAL IN UNITS OF ROOT (RESIDUAL GXE MS), ROWS AND COLUMNS SORDED ACCORDING TO MARGINAL MEANS)

		P I	T Y	N A	R O	L A	R I	A L	K L	M E	T - E F C T S
EB	EB	0	0	0	0	0	-1	0	0	0	EA
Y2	Y2	0	0	0	0	0	0	0	0	0	EA
EA	EA	0	0	-1	0	0	0	0	0	1	EA
H4	H4	0	0	0	0	0	1	0	0	0	EA
G1	G1	0	1	0	0	0	0	0	0	0	EA
H3	H3	1	0	0	0	0	1	0	0	0	EA
G2	G2	0	0	0	2	0	0	0	0	-1	EA
H6	H6	-1	0	-1	0	0	0	0	0	0	EA
H5	H5	0	0	0	0	-1	1	0	0	0	EA
H1	H1	-1	0	0	1	1	0	0	0	0	EA
YC	YC	2	0	0	-2	0	-1	0	0	0	EA
D3	D3	0	0	0	0	0	-1	0	0	0	EA
CA	CA	0	0	0	0	1	-1	-1	0	0	EA
H2	H2	0	0	0	1	0	0	1	-1	0	EA
D2	D2	-1	0	0	1	0	0	0	0	0	EA
D1	D1	0	0	1	0	0	0	0	0	0	EA
CD	CD	0	0	0	0	0	0	0	0	0	EA
CE	CE	0	0	0	0	0	0	0	0	0	EA
D4	D4	0	0	0	0	0	0	0	0	0	EA
CC	CC	0	0	0	0	0	0	0	0	0	EA

```
BOX PLOT OF  180 STANDERSIZED RESIDUALS FROM LPLT= -2.144    TO ULPT=  2.647
NO.<LPLT                                                          NO.>UPLT
       0 *  ------------------I    +     I--------------    *      *   0
```

ANALYSIS OF VARIANCE FOR THE AMMI MODEL

SOURCE	D.F.	S.S.	M.S.	F	FPROB
TREATMENTS	19	258.623	13.6117		
LOCATIONS	8	316.538	39.5672		
TREATMENT X SITES	152	269.669	1.77414		
AMMI COMPONENT 1	26	164.647	6.33257	7.597	0.000
AMMI COMPONENT 2	24	43.9112	1.82964	3.054	0.000
AMMI COMPONENT 3	22	21.6677	0.984896	1.998	0.014
AMMI COMPONENT 4	20	15.6286	0.781428	1.969	0.023
GXE RESIDUAL	60	23.8148			
TOTAL	179	844.830			

 GENOTYPE MAP SHOWING BEST GENOTYPES OVER THE RANGE OF AMMI-2 SITE SCORES (2 CHRS/PIXEL)

```
 1.37 CDCDCDCDCDCDCDCDCDCDCDCDCDCDEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBH6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6
 1.31 CDCDCDCDCDCDCDCDCDCDCDCDCDCDEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBH6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6
 1.25 CDCDCDCDCDCDCDCDCDCDCDCDCDCDEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBH6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6
 1.19 CDCDCDCDCDCDCDCDCDCDCDCDCDCDEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBH6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6
 1.14 CDCDCDCDCDCDCDCDCDCDCDCDCDCDEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBH6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6
 1.08 CDCDCDCDCDCDCDCDCDCDCDCDCDCDEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBH6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6H6
 1.02 CDCDCDCDCDCDCDCDCDCDCDCDCDCDEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEBEB
```

```
-0.71 CDCDCDEBEBEBEBEBEBEBEBEBEBY2Y2Y2Y
```

 SECTION 1

VARIETY\SITE		AL	KL	LA	ME	NA	PI	RI
EB	EB	0.7255	-.5186	0.4110	-.9182E-01	0.1536	0.5353	-1.157
Y2	Y2	-.2330	0.7227E-01	0.7119	0.2638	0.7544E-01	0.1618E-01	-.3107
EA	EA	0.6347	0.1408	-.3510	0.9705	-.7770	-.2488	0.6791
H4	H4	0.2119	-.2988	-.7151	-.5666	0.2178	0.2698	0.8523
G1	G1	-.5096	0.7275	-.7327	0.6379E-02	0.2087E-01	-.3108	-.3141
H3	H3	-.7302	-.1810	-.1471	-.3251	-.1886	1.038	0.9246
G2	G2	0.3799	-.3005	-.9734E-01	-.8304	0.7178	-.7487	-.1914
H6	H6	-.3904	0.3502	0.1741	0.5926	-.8552	-1.011	0.5065
H5	H5	-.4727	0.5613	-1.458	0.1266	-.2420	0.7313	1.312
H1	H1	0.2081E-01	-.4250	0.8209	-.1404	-.1179	-1.055	0.7421
YC	YC	-.5944	-.2472E-01	0.5970	-.8185E-01	0.6243	2.049 **	-.8878
D3	D3	0.3714E-01	0.3338	0.5145	0.2527	0.4824	-.5676E-01	-1.360
CA	CA	-.9779	0.4167	0.9930	-.1281	0.4620	-.3694	-1.048
H2	H2	1.129	-1.074	-.2180	-.7184	-.1466	-.2433	0.1990
D2	D2	0.5326E-01	-.8236E-01	0.1622	-.1596	-.1756E-01	-1.020	0.4124
D1	D1	-.2678	0.3026	-.3552	-.5847	0.8964	0.1533	-.6317
CD	CD	0.1778	-.1368	0.2447	0.3127	-.3100	0.1873E-01	0.1876
CE	CE	0.3764	-.6727E-01	-.4971	0.3428	-.4626	0.4353	0.3904
D4	D4	0.1486	0.2029	0.3340	0.4987	-.3082	-.7648	-.3140
CC	CC	0.2808	0.4710E-03	-.3918	0.2600	-.2249	0.5823	0.8950E-02
SITE EFFECTS		-.8597 ***	-1.826 ***	0.4035E-01	-1.893 ***	1.430 ***	1.707	-.7123 ***
AMMI1 SITE		0.1108 ***	-1.027	-.5365 ***	0.6407E-01	2.406 ***	-1.324 ***	1.018 ***
AMMI2 SITE		0.8695 ***	1.366 ***	0.4227 ***	-1.685	-.1945	-.3569	0.3908

SECTION 2

VARIETY\SITE		RO		TY		T-EFCTS		AMMI1 TRT		AMMI2 TRT	
EB	EB	-.5118		0.4543		2.392		0.1970		-.4672	
Y2	Y2	-.4760		-.1198		2.244	***	0.2692	***	-.7370	
EA	EA	-.6546		-.3937		1.207		0.2394		-.5385	***
H4	H4	0.7544		-.7256		1.133		-.9376E-01		0.7540E-01	
G1	G1	0.2679		0.8445		0.9848	***	-.2074		0.3233	
H3	H3	-.4500		0.5950E-01		0.7626		0.2239		0.1096E-01	
G2	G2	1.620	*	-.5489		0.4663		-.3149	***	-.8964E-01	
H6	H6	-.6861E-01		0.7019		0.3922		0.7147	***	0.7503	
H5	H5	-.2165		-.3423		0.2183E-01		-.1481E-01		-.1697	***
H1	H1	0.8619		-.7071		-.5224E-01		0.5359	***	0.4455E-01	
YC	YC	-1.660	*	-.2131E-01		-.1263	***	0.5988	***	0.8292	***
D3	D3	-.3952		0.1911		-.2004		0.4234	***	0.9772	***
CA	CA	-.1100E-01		0.6627		-.3485		-.8041	***	-1.362	
H2	H2	1.010		0.6190E-01		-.5708	***	0.2196	***	0.1731	
D2	D2	0.9735		-.3223		-.9411		0.3293		0.2891	***
D1	D1	0.6569		-.1698		-1.015		0.2330	***	-.1087E-01	
CD	CD	-.4018		-.9294E-01		-1.081		-3.144	***	0.7498	***
CE	CE	-.5672		0.4934E-01		-1.312		0.3335	***	-.4238	
D4	D4	-.9834E-01		0.3011		-1.682	***	0.4774	***	0.2458	
CC	CC	-.6331		0.1172		-2.274		-.2163	***	-.6698	
SITE EFFECTS		0.5737	***	1.540		73.46		.		.	
AMMI1 SITE		-1.505		0.7943	***	.		.		.	
AMMI2 SITE		-.8192		0.6439E-02		.		.		.	

FITTED VALUES FROM THE AMMI MODEL FILE 07_TSTW   16/ 7/ 9 16:37
 -- :PAGE 11

 SECTION 1

VARIETY\SITE		AL	KL	LA	ME	NA	PI	RI
EB	EB	74.61	73.19	75.59	74.76	77.85	77.46	75.16
Y2	Y2	74.23	72.59	75.29	75.07	77.92	77.32	74.98
EA	EA	73.37	71.86	74.35	73.70	76.78	76.25	73.99
H4	H4	73.79	72.97	74.72	72.57	75.78	76.40	73.81
G1	G1	73.84	73.27	74.73	71.99	75.31	76.31	73.65
H3	H3	73.40	72.18	74.15	72.33	76.19	75.63	73.74
G2	G2	72.95	72.30	74.10	72.16	74.62	76.08	72.86
H6	H6	73.72	72.32	73.83	70.74	76.86	74.34	74.16
H5	H5	72.47	71.44	73.46	71.87	74.91	75.27	72.69
H1	H1	72.65	71.09	73.18	71.47	76.12	74.39	73.26
YC	YC	73.26	72.02	73.40	70.08	76.04	73.95	73.55
D3	D3	73.30	72.33	73.49	69.75	75.52	74.06	73.36
CA	CA	70.98	70.25	73.01	73.46	72.87	76.37	71.05
H2	H2	72.20	71.07	72.88	70.72	74.81	74.24	72.47
D2	D2	71.95	70.75	72.50	70.16	74.68	73.69	72.25
D1	D1	71.60	70.36	72.36	70.58	74.44	73.85	71.97
CD	CD	71.82	74.80	74.42	69.02	66.10	77.98	68.76
CE	CE	70.96	69.40	71.83	70.99	74.46	73.56	71.61
D4	D4	71.18	69.80	71.67	69.50	74.31	72.76	71.65
CC	CC	69.72	68.67	71.06	70.41	72.22	73.42	69.99
SITE ESTS.		72.60	71.63	73.50	71.57	74.89	75.17	72.75
AMMI1 SITE		0.1108	-1.027	-.5365	0.6407E-01	2.406	-1.324	1.018
AMMI2 SITE		0.8695	1.366	0.4227	-1.685	-.1945	-.3569	0.3908

SECTION 2

VARIETY\SITE		RO	TY	T-ESTS.	AMMI1 TRT	AMMI2 TRT
EB	EB	76.51	77.55	75.85	0.1970	-.4672 I
Y2	Y2	76.48	77.45	75.70	0.2692	-.7370 I
EA	EA	75.32	76.39	74.67	0.2394	-.5385 I
H4	H4	75.25	76.06	74.59	-.9376E-01	0.7540E-01 I
G1	G1	75.07	75.82	74.44	-.2074	0.3233 I
H3	H3	74.45	75.94	74.22	0.2239	0.1096E-01 I
G2	G2	75.05	75.22	73.93	-.3149	-.8964E-01 I
H6	H6	72.74	75.96	73.85	0.7147	0.7503 I
H5	H5	74.22	75.01	73.48	-.1481E-01	-.1697 I
H1	H1	73.14	75.37	73.41	0.5359	0.4455E-01 I
YC	YC	72.33	75.35	73.33	0.5988	0.8292 I
D3	D3	72.40	75.14	73.26	0.4234	0.9772 I
CA	CA	76.01	74.00	73.11	-.8041	-1.362 I
H2	H2	72.99	74.60	72.89	0.2196	0.1731 I
D2	D2	72.36	74.32	72.52	0.3293	0.2891 I
D1	D1	72.68	74.17	72.44	0.2330	-.1087E-01 I
CD	CD	77.07	71.43	72.38	-3.144	0.7498 I
CE	CE	72.57	73.95	72.15	0.3335	-.4238 I
D4	D4	71.43	73.70	71.78	0.4774	0.2458 I
CC	CC	72.63	72.55	71.19	-.2163	-.6698 I
SITE ESTS.		74.03	75.00	73.46	.	.
AMMI1 SITE		-1.505	0.7943	.	.	.
AMMI2 SITE		-.8192 I	0.6439E-02 I	.	.	.